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CHLORPROPAMIDE AND DIABETES MELLITUS*

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PREFACE

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A new era in the field of diabetes mellitus has been ushered in with the introduction of effective oral hypoglycemic agents particularly the sulfonyl ureas The great impact made by these agents was evident in the success attending the conference on *Sulfonylureas and Related Compounds* sponsored by The New York Academy of Sciences in February 1957 It was apparent from this conference that compounds such as tolbutamide are to be added to the armamentarium useful in the management of diabetes These substances appear capable of controlling the condition in adult onset diabetes with

only because of their use in the treatment of diabetic states but also because they served to rekindle interest and further research in the etiology of diabetes in the mechanisms of insulin synthesis storage and release and in the role of the liver and peripheral tissues in carbohydrate metabolism

The present conference centers about sulfonylurea derivatives particularly chlorpropamide It is becoming increasingly evident that incorporation of members of the halogen series into the structure of organic molecules often enhances their biological activities This is true of hormones of adrenocorti cal origin in which for example substitution of halogens such as fluorine into the 9- and 6 alpha positions results in considerable augmentation of certain physiological actions A similar situation is seen in the synthesis of thyronine derivatives in which iodine is superior to bromine and chlo ride This concept also extends to the sulfonylurea compounds notably chlorpropamide

We look forward to further detailed information on topics relating to the mechanism of action of the sulfonylureas particularly their influence on insulin metabolism to a comparison of the relative effectiveness and toxicity of the newer derivatives especially chlorpropamide and to the extension of their use in the treatment of diabetic states in man All of these topics are discussed in this monograph

INTRODUCTORY REMARKS

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It is my hope that the presentations and discussions comprising this monograph will bring us another step forward in the understanding of the role that the hypoglycemic sulfonylurea derivatives can play in the management of diabetes mellitus

When in 1942 the first chance observations were made at the University of Montpellier in southern France about the hypoglycemic action of a new sulfonamide, Loubatières foresaw that at last the long and so far rather fruitless search for an oral drug for the treatment of diabetes mellitus had reached a new phase and that a promising agent was finally at hand

However, it took another chance observation, this time by German investigators, and another decade until the first such drugs—tolbutamide and carbutamide—could be introduced into the practice of medicine During the past few years nearly 1000 other sulfonylurea derivatives have been developed and evaluated in various laboratories Chlorpropamide, or Diabinese, is one of these compounds Because of such special characteristics of this preparation as its effectiveness in rather small doses and its lasting action, it was given an extended clinical tryout The reports of these trials included in these pages will give us an opportunity to decide whether a new and practically useful agent has come into our hands in addition to those presently available

Part I Pharmacological Studies

GENERAL PHARMACODYNAMICS OF THE HYPOGLYCEMIC ARYLSULFONAMIDES

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The question of the general pharmacodynamics of the hypoglycemic arylsulfonamides of which the following* is a brief survey has been the subject of a number of our more deta led articles [1-10] We shall discuss here only the essential points in regard to this important question

First we should record the fact that the basic experimental discover es made in our laboratory between 1942 and the present time have been con

3 *p* tolylsulfonylurea (D 860 tolbutamide)

The compound 1 propyl 3 (*p* chlorobenzenesulfonyl)urea (P 607) chlorpropamide is still under study and at present some reservations must be expressed regarding it Although its mechanism of action is probably the same as that of the compounds mentioned above this is not established also we are not well informed as to its toxicity and tolerance It is the purpose of

sulfonamide or arylsulfonamide as seems suitable

Mechanism of Action Experimental Study

The fundamental and elemental role played by the endocr ne pancreas (the beta cells of the islets of Langerhans) and insulin characterizes the mechanism of action of the arylsulfonam des

As long ago as 1944 we demonstrated that the sulfonamides do not produce hypoglycemia in the totally depancreatized dog but that if even a fraction of the pancreas containing some islets of Langerhans is left in place the hypoglycemic action of these substances is man fested When these substances are introduced into the organism they come into contact by way of the blood stream with the islets of Langerhans for which they have a special tropism they are pancreatropic

They act as stimulants to the beta cells from which they release a certain amount of endogenous insulin into the blood Numerous experimental deductions support this statement

* Th s account is a rev se l vers on of that presented at the meet ng of the Th d International Congress of the Internat onal D abetes Federat on held from July 21 to 25 1958 at Düssel lorf Germany

The effects of these compounds are as follows (1) the beta cells lose some of their intraprotoplasmic insulin granules, (2) the insulin content of the pancreatic tissue diminishes in response to the development of glycemia, (3) as shown by us in 1946 by means of experimental pancreatic *jugular anastomosis*, the hypoglycemic activity of the blood leaving the pancreas *increases* considerably a fact since confirmed in several ways, especially by measuring the insulin activity of the blood plasma leaving the pancreas

Recently we resumed these experiments and showed that the venous [illegible]

before the action of the drug

In se[illegible]

the[illegible]

ate[illegible]

sulfamides are hypoglycemic

It is also known that several days of sulfonamide administration produce in the normal animal a hyperplasia of the islet beta cell system and stimulate the formation of new beta cells that come into being at the expense of cells of the exocrine pancreatic gland (the centroacinar cells, cells of the excretory canaliculi the acinar cells) Here is a demonstration of the validity of Laguesse s theory that the cells of the exocrine gland may be the origin of endocrine cellular elements

In 1956 we studied the roles of the endocrine glands other than the pancreas in the mechanism of action of the arylsulfonamides The following experiments carried out in our laboratory demonstrate the fundamental role of the pancreas

Using the same dog, we removed, in several operative procedures, the following endocrine glands, one after another, in this order (1) the two adrenals (cortical and medullary tissues) (2) the thyroid, the four parathyroids, and the genital glands, and (3) the entire hypophysis

After each of the operative procedures the sulfonamide proved to be hypoglycemic, except following the removal of the entire pancreas However, it is clear that hypophysectomy and, especially, adrenalectomy sensitize the organism to the hypoglycemia induced by the sulfamides The sulfonamides retain their hypoglycemic action in the hepatectomized animal, provided the pancreas is intact However, this does not imply that the liver plays no role

The glycogenic action that these compounds exert upon the liver, demonstrated by us in 1944, is the basis for postulating that the sulfonamides act by inhibiting certain enzymes (glucose 6 phosphatase, reactivation of phosphorylase) However, these effects do not seem to be specific and are manifest only at high concentrations of the drugs Moreover, the inhibition they produce is only partial

Our recent experiments upon the dog subjected to a preliminary eight-day fast, which slows insulin secretion, show clearly that liver glycogen formation can be explained by the effects on the liver cell of endogenous insulin released by the sulfonamide Furthermore, we observed that administration of

sulfonamide also induces an accumulation of glycogen in muscle, that is to say it affects the peripheral tissues Both of these results the hepatic and the muscular, of the sulfonamides may be reproduced completely by

depancreatized dog is preserved completely These facts show that the sulfonamides do not seem to inhibit profoundly the enzymatic reactions involved in glycogenolysis or hepatic glycogen formation in the physiological conditions under which we operated

Furthermore, we showed at the same time as Houssay and his collaborators that the sulfonamides in massive doses potentiate the hypoglycemic effects of insulin even after ablation of the pancreas It has not been demonstrated that this potentiation of the effects of insulin is due to an inhibition of insulin ase We have other hypotheses Whatever the explanation the release of endogenous insulin precedes the subsequent intensification of the effects of this hormone A possible action of the sulfonamides on the peripheral tissues in the absence of insulin or the pancreas has not been demonstrated Recent study of the peripheral arteriovenous differences in blood sugar after sul fonamide and after insulin shows striking similarities

On the other hand we have shown that the administration of sulfonamide is capable of accumulating glycogen in the striated muscle of a dog subjected to prolonged preliminary fasting We have reason to believe that endoge nous insulin released by the sulfonamide and not wholly arrested by the hepatic barrier is responsible for these peripheral effects

In addition to their hypoglycemic action the sulfonamides possess an antidiabetic action that is exhibited under certain experimental conditions in the dog with alloxan diabetes We have shown that, if the diabetes is severe (that is to say, if all of the beta cells have been destroyed by the alloxan) the sulfonamides are inactive, they may even hasten the evolution of the diabetes toward acidosis

On the other hand, if the diabetes is of moderate intensity, the action of the sulfonamides is evident The different manifestations of the diabetes progessively diminish and then disappear After several weeks the treatment can be discontinued, and the animal is "cured " New beta cells have devel oped in the pancreas of these animals We have shown that these cells are functionally normal, since they are perfectly capable of regulating the blood sugar, they are therefore sensitive to the delicate stimuli for insulin secretion They can be stimulated by the sulfonamides and, in turn, they can be destroyed by alloxan

This new islet apparatus whose functional activity exists thanks to the

gressively hyperglycemic by the end of six months This shows the impor

tance of a controlled diet for maintaining the benefits obtained from the sulfonamides

Mechanism of Action in Man

Can proof of hypoglycemic antidiabetic action be established in normal or diabetic man? The hypoglycemic action of the sulfonamides is admitted without reservation in the case of normal man On *rare occasions these substances cause serious hypoglycemic accidents that, in any case, are reversible* by glucose In a man totally without pancreatic tissue these substances are inactive, they maintain their activity if a small amount of endocrine pancreas remains in the organism

[illegible]

sugar The effects observed depend, therefore, on the respective and oppo site roles played in establishing the total diabetic state by the pancreatic component, on the one hand and the extrapancreatic diabetogenic component on the other *This explains the variable results observed following the use of* sulfonamides when glandular hyperfunction (anterior pituitary or adrenal cortical) is beginning, or when this hyperfunction is being reduced The hemosiderin depot observed in the beta cells of the islets of Langerhans probably is not unrelated to the lack of action of the sulfonamides in hemochromatosis (bronze diabetes) This behavior confirms once more the tropism of the sulfonamides for the beta cells of the pancreas

In cirrhosis of the liver with diabetes the activity of the sulfonamides is variable and depends on many factors In the diabetic man under insulin [illegible]

[illegible] sulfonamide added its effects to that of the injected insulin? Do other mechanisms enter in? These questions are still unanswered

We have recently answered several questions in regard to sulfamide treatment It has been stated, for example that the sulfonamides are not physiological agents That is true, but their pharmacological action causes endoge [illegible]

explained by the degree of lesion of the beta islet cells of the pancreas [illegible] different stages that represent the various degrees of intensity of the diabetes

The classification of diabetic subjects in terms of their different responses to the sulfamides is a strong argument in favor of our theory of the mechanism of action of these substances Young diabetics, refractory to the *sulfamides,*

have severe beta cell lesions in their pancreas, the insulin content of this organ is low, the stimulation of its cells by glucose administration releases practically no detectable insulin into the blood

On the other hand, diabetics stricken with the disease at a mature age have numerous intact beta cells in their pancreas, the insulin content of this

acknowledged that the sulfonamides seem to act especially in subjects exhibit ing a certain degree of sluggishness or insufficiency of the insulin secretory processes Despite the considerable amount of work accomplished since 1946, the theory that emerges remains in conformity with the one we have maintained since then and will continue to maintain, because it is well founded

There is a question as to whether the chronic stimulation of the beta cells may not finally bring on their exhaustion and an aggravation of the diabetes Taking into consideration our long experience, it does not seem that this contingency often arises when the sulfonamides are correctly used in subjects who are really suitable When this condition arises, it is necessary to take into account deviations from the diet, the way in which the sulfonamides are used, and the spontaneous aggravative evolution of certain cases of dia betes before the sulfamides themselves can be held entirely responsible Conditions called refractory to the sulfonamides may appear, they may be explained in part by the phenomenon of tachyphylaxis

Hypoglycemic Sulfamides and the Prospects for Extended Remission of Diabetes Mellitus

In conclusion we should like to explain why, in our opinion, the hypo glycemic and antidiabetic sulfamides offer favorable prospects for pro phylaxis or extended remission of diabetes

Accumulated clinical data show two types of results in certain elderly diabetics or even in young diabetics (provided their diabetes is recent and not severe) a sulfamide treatment carried on for several weeks can cause the diabetes to disappear In many cases the benefits last for several months after suspension of sulfamide treatment The sulfamides have, therefore, an antidiabetic action in man as well as in animals

It is possible that the extended remission of diabetes, which is not a final cure, is explained by at least partial regeneration of the endocrine pancreatic system If this is true why not attempt to use the sulfonamides as pro phylactic and retarding agents in subjects predisposed to this disease?

There is another point that should engage our attention, since it presents a problem and indicates a line of research It involves the observation of

the cured' animal the blood sugar is normal and, if the diet is correctly controlled, the 'cure" lasts Lesions caused by alloxan were pancreatic,

the sulfonamides have compensated for the pancreatic lesions at tleas in part by facilitating the regeneration of the beta cells, *glycemic equilibrium* has returned and will be prolonged if certain experimental conditions are respected In extended remission of diabetes in man, blood sugar is almost never normal but is definitely above 1 10 to 1 20 gm of glucose per liter After several weeks or months the diabetes progressively resumes with all its former intensity What has happened?

The sulfamide treatment it is true has partially restored the endocrine

of diabetes

Thanks to the research accomplished in the domain of the hypoglycemic sulfonamides an important advance has been made, but our task as seekers is not yet over Now we must undertake to detect and neutralize the harmful extrapancreatic influences It is probable that we shall not be too long without weapons against these factors We should succeed in this because these influences often are exerted only temporarily in the course of a lifetime (period of somatic growth premenopausal period), and science should not lose the hope of overcoming them once they are detected

The "cure of experimental diabetes is possible if the disease has not exceeded a certain degree of severity It is not unreasonable to believe that we are on the way to realizing within organically permitted limits at least, the extended remission of human diabetes mellitus If we arrive at this result the challenge offered by the hypoglycemic and antidiabetic sulfonamides sixteen years ago to diabetologists will not, from the scientific, the social and the human points of view have been in vain

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STUDIES ON THE EFFECT OF ORAL ANTIDIABETIC COMPOUNDS ON GLUCOSE TOLERANCE, ON ISLET CELL STRUCTURE, AND ON THE *IN VITRO* METABOLISM OF ISOLATED MUSCLE

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The mode of action and the locus or loci of action of the individual oral antidiabetic compounds have not been fully elucidated. We shall present briefly the results of three kinds of studies in which we have tried to obtain some information about the action of these compounds.

Oral and Intravenous Glucose Tolerance During the Action of Oral Antidiabetic Compounds

The first study is of glucose tolerance in diabetic patients.[1] FIGURE 1 shows the average oral glucose tolerance curve before and during the administration of carbutamide or tolbutamide in 18 patients. The antidiabetic effect is

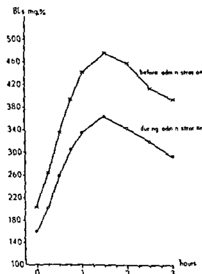


FIGURE 1. Average curves showing oral glucose tolerance before and during the administration of carbutamide or tolbutamide.

quite obvious. It is not only a parallel displacement of the curve that occurs, the area of increase, that is, the area above fasting level, is significantly decreased during the action of the sulfonylureas. FIGURE 2 shows the results of intravenous glucose tolerance tests. The individual k values,

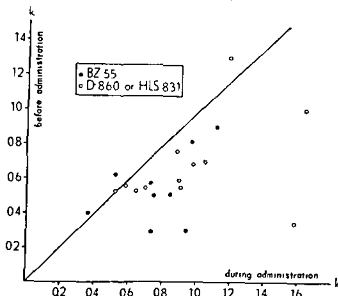


FIGURE 2 *k* Values for intravenous glucose tolerance curves before and during the administration of carbutamide (BZ 55) or tolbutamide (D 860 or HLS 831)

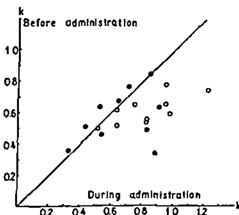


FIGURE 3 *k* Values for intravenous glucose tolerance curves before and during the administration of chlorpropamide Symbols ● = 1 75 gm during a period of 48 hours ○ = 3 to 5 gm during a period of 48 to 96 hours

expressing the slope of the decrease in blood sugar in each of the patients before and during treatment with sulfonylureas, are plotted against each other An antidiabetic effect on the intravenous glucose tolerance is seen, the points clearly tending to accumulate below the 45° identity line

sugar[2] Even by these devious ways we find that an antidiabetic effect although less significant, can be shown to occur in the intravenous glucose tolerance test Baird and Duncan[3] have published findings very similar to ours but, by using the blood sugar excess values they failed to demonstrate any significant effect

TABLE 1 shows the serum inorganic phosphate during glucose tolerance tests before and during the action of sulfonylureas It appears that the

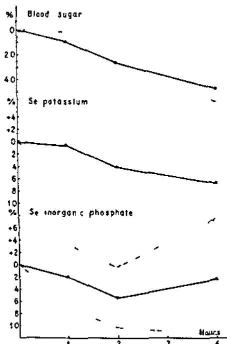


FIGURE 4 Blood sugar serum potassium and serum inorganic phosphate 1 2 and 4 hours after oral administration of 1 gm chlorpropamide Values are shown as per cent changes from the before values Dotted lines represent standard deviation

amelioration of the glucose tolerance was not accompanied by any change in the phosphate response

FIGURE 3 shows the results of intravenous glucose tolerance tests in 20 diabetic patients before and during administration of chlorpropamide It appears that 1 gm /day of this compound produces an amelioration of the glucose tolerance similar to the one found with carbutamide or tolbutamide However chlorpropamide increased to some extent the response of the inorganic serum phosphate to glucose (TABLE 1) and when given in the fasting state it produced a small but significant fall of serum phosphate and of serum potassium (FIGURE 4)

TABLE 1

AVERAGE SERUM INORGANIC PHOSPHATE DURING ORAL AND INTRAVENOUS GLUCOSE TOLERANCE TEST BEFORE AND DURING THE ADMINISTRATION OF CARBUTAMIDE (C), TOLBUTAMIDE (T) OR CHLORPROPAMIDE (CP)

Glucose tolerance test	Antidiabetic compound	No of patients	Time after oral or I V glucose (min)				Max fall	Percentage fall
			0	60	120	180		
Oral	0	18	3 8	3 6	3 5	3 4	0 41	10 8
Oral	C or T	18	3 7	3 4	3 3	3 4	0 41	11 1
Intravenous	0	21	3 7	3 5				5 4
Intravenous	C or T	21	3 6	3 4				5 6
Intravenous	0	20	3 6	3 5				3 8
Intravenous	CP	20	3 8	3 5				7 1

It is not clear if this difference between the response of the inorganic phosphate to carbutamide and tolbutamide, on the one hand, and to chlorpropamide, on the other, is due to a difference in the mode of action of these compounds It should be noted that under certain circumstances a fall of serum inorganic phosphate has been reported to occur after administration of tolbutamide [4, 5]

Pancreatic Islets of the Rat During the Administration of Sulfonylureas

The second type of study concerns the histological structure of the pancreatic islets in the rat during the administration of sulfonylureas [6]

Normal female rats were injected intraperitoneally with p aminobenzenesulfonamidoisopropylthiodiazole (IPTD), carbutamide, or tolbutamide in

experi

The islet

ollowing

the details of the procedure described by Gomori [7] The results are shown in TABLE 2 No difference in the structure or granulation of the alpha or beta cells of the controls or of the injected animals was obtained Synthalin A

tion in the rat They differ in the same respect from the results of studies in the calf[9] and in the rabbit [10] This has been interpreted as histological support for the hypothesis that the oral antidiabetic compounds act by increasing the production and/or output of insulin from the pancreatic islets In our laboratory, working with rats we have not been able to produce any histological evidence for this hypothesis

However we emphasize that we have had considerable difficulty with the Gomori stainings, which seem to vary somewhat from day to day, giving

TABLE 2
HISTOLOGICAL CHANGES IN PANCREATIC ALPHA AND BETA CELLS OF RATS AFTER ADMINISTRATION OF VARIOUS ANTIDIABETIC DRUGS

Drug	No of animals	No of controls	Total dose (mg)	Histological changes of	
				Alpha cells	Beta cells
Carbutamide	75	29	75–33 5	0	0
Tolbutamide	44	11	75–3375	0	0
IPTD	28	9	75–3375	0	0
Synthalin A	10	3	20	+++	0
Total	157	52	—	—	—

the illusion of a change in the granulation of the beta cells We have found that it was always necessary to carry the control specimens in the same run all the way through the entire procedure

In Vitro Studies of Isolated Rat Diaphragm

The third type of experiment deals with the action of sulfonylureas on the isolated diaphragm of the fasting rat [11]

For these experiments the usual Gemmil technique was used The medium was Gey and Gey's buffer with 300 mg per cent of glucose The antidiabetic compounds were either injected into the animals a few hours before the experiment or were added to the medium Each experiment included 6 to 12 treated and 6 to 12 control hemidiaphragms In injection experiments we attempted to produce a blood level of about 10 to 50 mg per cent of carbutamide In additional experiments the medium contained from 6 to 100 mg per cent of the antidiabetic compound

In injection experiments with carbutamide and in experiments in which carbutamide tolbutamide IPTD, or phenethyldiguanide was added to the medium, a significant rise in the glucose uptake was observed consistently No change occurred in the glycogen content of the muscle except with phenethyldiguanide, which produced a fall in the muscle glycogen

The results of the experiments with antidiabetic compounds added to the medium are summarized in FIGURES 5 and 6

Our findings demonstrate that IPTD carbutamide, and tolbutamide can increase the uptake of glucose in the isolated muscle tissue without affecting glycogen deposition that is, changes in the metabolism of the muscle that differ from the action of insulin on isolated muscle tissue Similar results have been obtained by Canal *et al* [12] and by Pletscher and Gey,[13] but other investigators have reported negative results of such studies [14 17] Our results, using phenethyldiguanide, are in accordance with the results reported by Williams *et al* [18]

The results obtained in our laboratory indicate that the action of the

antidiabetic compounds on the muscle is independent of the presence of active beta cells that it is not brought about by an increase in the production and delivery of insulin from the pancreas

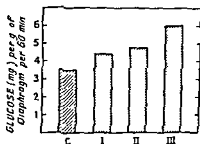


FIGURE 5 Average uptake of glucose

I carbutamide, II tolbutamide, III phenethyldiguanide concentration 35 mg per cent The difference between each of these average values is statistically significant

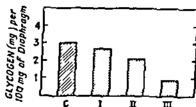


FIGURE 6 Average glycogen content of the isolated rat diaphragm after incubation in glucose-containing medium with various oral antidiabetic compounds C, I, II, III and concentration are as in FIGURE 5 III is significantly lower than C I, and II

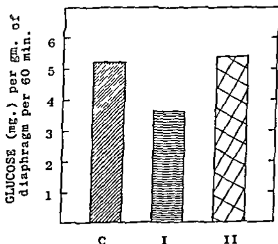


FIGURE 7 Average uptake of glucose by the isolated diaphragm of alloxan diabetic rats C, controls I alloxan diabetic rats, II alloxan-diabetic rats (concentration of carbutamide in the medium is 35 mg per cent) C and II are significantly higher than I

We have studied also the effect of carbutamide on the glucose uptake of diaphragms isolated from alloxanized rats The rats were moderately to ... after the injection of alloxan [19]

the same glycogen

content (FIGURE 7)

Of course these results are in disagreement with the hypothesis that the action of the oral antidiabetic compounds require the presence of beta cells capable of increasing the output of insulin However it is not possible to prove that an alloxanized animal is an insulin free organism and we are well aware of the fact that our results with alloxanized rats do not disprove the hypothesis supported by many other kinds of evidence that the action of these compounds requires the presence of insulin We do believe that it is important to distinguish the hypothesis of action by increased insulin secretion from the hypothesis of insulin as a permissive agent for the action of the oral antidiabetic compounds

There is good evidence from several sources[20 22] that the oral antidiabetic compounds reduce the output of glucose from the liver but it is known that they are active even in the absence of this organ [15 23] The results presented here suggest that an action on the peripheral uptake of glucose might also contribute to the fall of blood sugar observed after administration of these compounds

Discussion

We did not measure oxygen uptake in these experiments but I do not think that lack of oxygen can explain the increase of glucose uptake observed in our experiments with oral antidiabetic compounds We are working with a bicarbonate phosphate buffer and an atmosphere of carbogen In our control experiments with nonhypoglycemic sulfonylureas we did not find any increase in glucose uptake Our system is highly sensitive to insulin both on glucose uptake and on glycogen deposition

There is some evidence of a more general enzyme inhibition with very high concentrations of sulfonylureas A lack of increase of glucose uptake by the isolated diaphragm with such high concentrations has also been reported from other laboratories

Summary

In clinical studies of the oral and intravenous glucose tolerance an increase of glucose tolerance was observed during the administration of carbutamide tolbutamide or chlorpropamide to diabetic patients Serum inorganic phosphate was unchanged during the action of carbutamide and tolbutamide With chlorpropamide a slight decrease in serum inorganic phosphate and in serum potassium occurred

Histological studies of the pancreatic islets of the rat showed no changes in the alpha or beta cells during the administration of low or high doses of IPTD carbutamide or tolbutamide for short or long periods of time

In vitro studies of the isolated rat diaphragm showed a rise in the glucose uptake when carbutamide was injected previously a similar rise was observed on the addition of IPTD carbutamide or tolbutamide to the medium *in vitro* No change in the glycogen content of the diaphragm was observed The same results were obtained using diaphragms from alloxanized rats with moderate or severe diabetes Phenethyldiguanide caused a rise in

the glucose uptake and a fall in the glycogen content of the isolated rat diaphragm

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THE PHARMACOLOGY OF CHLORPROPAMIDE

Jurg A Schneider Ernesto D Salgado Doris Jaeger Charles Delahunt
Department of Macrobiology Research Pfizer Therapeutic Institute Maywood N J

Ever since it was recognized that insulin could not be used orally in the treatment of diabetes mellitus both synthetic compounds and natural products have been investigated for hypoglycemic activity pharmacologically and clinically Severe side effects and/or insufficient activity made many of the previous attempts futile or impractical

The recent extensive studies of certain sulfonylureas as orally active hypo glycemic agents[1 2] are the result of a concentrated research effort of the last sixteen years[3] When the therapeutic value of the sulfonylureas was recog nized the search for the most effective compounds of the series was inten sified Chlorpropamide was selected as a result of an extensive screening program Its high degree of hypoglycemic activity and prolonged duration of action as evidenced by animal experimentation warranted thorough pharmacological and clinical studies[4]

This paper summarizes the present status of pharmacological investigation with chlorpropamide taking into account previous reports by Salgado *et al*[5] and Root[6] Emphasis is placed on new aspects data corroborating earlier findings are referred to briefly

Materials and Methods

The results reported were established in mice (Webster strain) rats (Wistar strain) cats dogs and rhesus monkeys Routine pharmacological and biochemical methods were employed unless otherwise indicated

All blood sugar determinations were performed according to the method of Folin and Malmros[7] The animals were fasted for 18 hours previous to the withdrawal of blood samples Liver and muscle glycogen were determined by the method of Seifter[8] Chlorpropamide blood levels were evaluated by a spectrophotometric method developed by Toolan and Wagner[9]

Chlorpropamide was administered orally in most experiments Where parenteral administration was employed the sodium salt of chlorpropamide was used (solubility in water > 50 per cent pH of 1 per cent solution = 8 5) Radioactive material consisted of S^{35} labeled chlorpropamide

Results

Hypoglycemic activity of chlorpropamide As reported previously,[5 6] chlorpropamide was found to cause a prolonged hypoglycemic effect in both rats and dogs Single oral doses of 25 mg /kg or higher were required to obtain a definite effect Calculated on a mg /kg basis, this is at least twice the recommended average human dose (7 to 10 mg /kg /day) When compared to tolbutamide in these two species chlorpropamide was somewhat slower in its onset but more prolonged in its duration of action, the maximal hypoglycemic effect of both compounds was of approximately equal magnitude

Root[6] had observed that, using both duration of action and degree of hypo glycemia as criteria for evaluation, in both rats and dogs chlorpropamide was more active on a weight basis than tolbutamide. The hypoglycemic effects of chlorpropamide and tolbutamide were also compared in rhesus monkeys following single oral administration. The results of such an experiment are shown in FIGURE 1. At dose levels of 10 mg/kg orally a definite hypoglycemic response was obtained that was slower in onset but considerably greater in magnitude and more prolonged in its duration of

HYPOGLYCEMIC ACTIVITY OF CHLORPROPAMIDE IN THE MONKEY

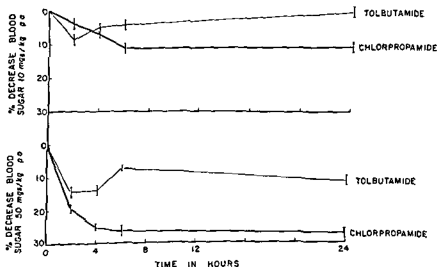


FIGURE 1. *Top tracing*: crossover studies in 4 monkeys illustrating the hypoglycemic effect of chlorpropamide as comp

action than with tolbutamide. At doses of 5 mg/kg orally (not shown on FIGURE 1) a definite blood sugar lowering effect was observed with chlor propamide whereas tolbutamide caused a slight and rather transient rise of blood sugar levels at that dose. At 50 mg/kg, which is an excessive dose, both compounds caused pronounced hypoglycemia, chlorpropamide again being more active and more prolonged in its effect than tolbutamide. There fore in the rhesus monkey chlorpropamide was active in dose levels that compare well to those used in the human being.[10] The results also indicate that rhesus monkeys, much like human beings, are more susceptible to chlorpropamide than either dogs or rats.

In view of the long duration of action of chlorpropamide, the question arose whether chlorpropamide had cumulative effects To clarify this point, groups of four dogs were given doses of 5, 25, and 50 mg/kg of chlorpropamide orally for several weeks Blood sugar and chlorpropamide blood levels were determined at regular intervals The results of this study are shown in FIGURE 2 It can be concluded that continued administration of subthreshold doses of chlorpropamide did not result in a hypoglycemic effect, even after 2 weeks of daily medication However larger doses caused a gradual decrease of the blood sugar that was paralleled by a rise in serum

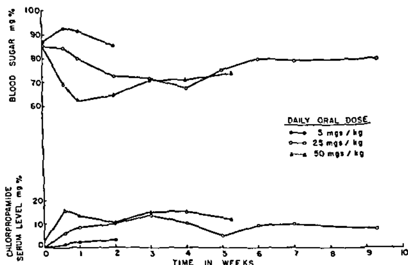


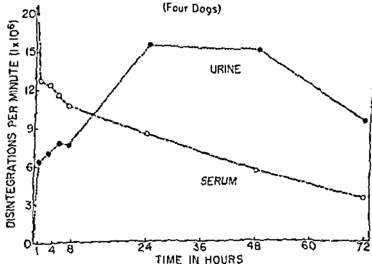
FIGURE 2 Comparison of hypoglycemic activity and blood levels of chlorpropamide in groups of 4 dogs following daily oral administration at various dose levels Each point represents an average of 4 values All blood sugar determinations and chlorpropamide blood levels were made 24 hours following the last dose, the animals were in a fasting state

concentration of chlorpropamide After several days the blood levels of chlorpropamide stabilized and did not increase beyond a certain point As noted by Root,[6] hypoglycemic effect and drug concentration in serum were not always in good correlation Reduction or cessation of medication resulted in gradual reduction of chlorpropamide blood levels and return of blood sugar to normal values

Since the method of Toolan and Wagner,[9] is not applicable for determination of chlorpropamide in urine, excretion studies were performed in dogs with radioactive S^{35} labeled chlorpropamide given intravenously The results available are summarized in FIGURE 3 According to these data the radioactive material is excreted very slowly, only about 45 to 50 per cent of the material given being accounted for after 72 hours Chromatographic

studies are in progress to determine whether chlorpropamide is excreted unchanged in the dog or whether it is metabolized

Toxicology In acute oral, intravenous, and intraperitoneal toxicity studies in mice, rats, and dogs, chlorpropamide was slightly more toxic than tolbutamide on a weight basis (TABLE 1), the species used, the age, and general status of the animal were important factors for survival following single doses This finding is in accord with the more potent hypoglycemic activity of chlorpropamide over tolbutamide



Chronic toxicity experiments were performed in dogs, rats, and monkeys at various dose levels for periods ranging from 6 to 20 months Dogs tolerated daily oral doses of 150 mg/kg, which corresponds to more than 20 times the recommended clinical dose in the human being Except for

administration Toxic symptoms observed were ataxia,
tremors, muscular weakness coma, and death

Rats survived daily doses up to 500 mg/kg in their diet for 6 months to 1 year A most interesting phenomenon developed in this species only at the high dose levels (FIGURE 4) Chlorpropamide caused a retardation in

TABLE 1
ACUTE TOXICITY OF CHLORPROPAMIDE AND TOLBUTAMIDE

Drug	Species	Route	LD_{50}	95% Confidence limits* mg/kg
Chlorpropamide	Mice	Oral	1675	1444-1943
Chlorpropamide	Mice	S C	780	710-856
Chlorpropamide	Mice	I P	760	693-834
Chlorpropamide	Mice	I V	500	431 580
Chlorpropamide	Rats	Oral	2390	1943 2940
Chlorpropamide	Rats	S C	760	603-958
Chlorpropamide	Rats	I P	575	454 725
Chlorpropamide	Rats	I V	590	486-714
Chlorpropamide	Dogs	Oral	approx 800	
Chlorpropamide	Dogs	I V	approx 575	
Tolbutamide	Mice	Oral	1830	1591 2105
Tolbutamide	Mice	I V	700	669-731
Tolbutamide	Rats	Oral	2490	2029-2936
Tolbutamide	Rats	I V	775	731 817

* Whenever possible LD_{50} and 95 per cent confidence limits were calculated according to the method of Litchfield and Wilcoxon CMC (carboxymethylcellulose) was employed as a suspending agent Rats and mice used were weanlings

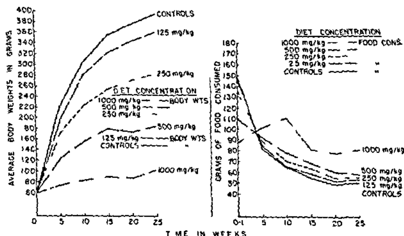
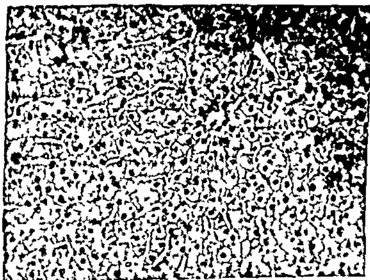


FIGURE 4 Growth-depressant effects of chlorpropamide in rats as evidenced by plotting average body weights of groups of twenty animals versus time (left graph) A *dose response effect* could be established clearly Feed consumption (gm /kg) in the corresponding groups (right graph) indicates adequate food intake

growth that was dose dependent the food intake of these rats however di not decrease proportionally In order to check whether chlorpropamid interfered with the availability of certain amino acids essential for growth cysteine and/or glycine were added to the diet in equimolar concentration t chlorpropamide Although glycine but not cysteine alleviated growth depression slightly these dietary adjustments failed to overcome chlorprop amide induced growth retardation

Rhesus monkeys are presently being maintained on chlorpropamide orally in doses up to 200 mg /kg (about 30 times the blood sugar lowering dose) intermittent diarrhea in the high dose range is the only toxic symptom observable



n from a dog treated orally No s gns of

Autopsies were performed on all animals sacrificed after periods of up to n was given to the i dog treated for As in all other anges This is in accord with the unimpaired liver function of [illegible] as evidenced by periodical liver function tests such as for alkaline phosphatase thymol turbidity or Bromsulphalein (BSP)

Mode of action Despite thorough pharmacological investigation qualitative differences between the mode of action of chlorpropamide and tolbutamide could not be established Chlorpropamide showed a marked effect on glycogen metabolism as evidenced by increased glycogen content of the liver (FIGURE 6) on the other hand glycogen content of striated muscle was

unaltered Insulin sensitivity was not increased by chlorpropamide (FIGURE 7), while glucose tolerance remained within the limits of normal In our laboratories similar results were observed with tolbutamide In accord with

several days (FIGURE 8) The phenomenon was reversible after discontinuation of medication for 72 hours Furthermore alloxan diabetic rats showed no hypoglycemic reaction to chlorpropamide, whereas severe hypoglycemia

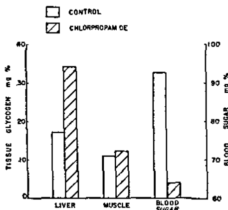


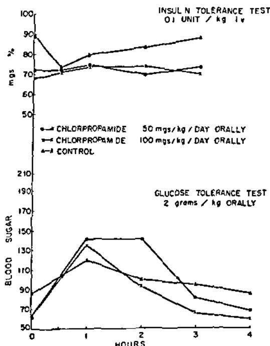
FIGURE 6 Glycogen contents in liver and muscle before and following chlorpropamide treatment in rats Six rats were used for each determination at a dose level of 100 mg kg orally The animals were sacrificed 2 hours after treatment the blood sugar determinations taken simultaneously indicate the effectiveness of the medication The animals were fasted throughout the test Chlorpropamide caused an increase in liver glycogen over the control but caused no change in muscle glycogen This is in accord with data in the literature obtained with other sulfonylureas

and death followed drug treatment of adrenalectomized rats (FIGURE 9) Similar experimental data are reported for tolbutamide [1, 2]

Chlorpropamide was also investigated thoroughly for pharmacodynamic actions other than those related to its hypoglycemic activity It lacked specific blood pressure, respiratory, or diuretic effects in doses that caused profound hypoglycemia There was no antagonism to histamine or acetylcholine in the isolated guinea pig ileum

The most obvious symptoms following high doses of chlorpropamide in mice rats, cats, and dogs are general weakness, associated with ataxia, and incoordination They may occur within a few minutes after intravenous

administration of high doses to an unanesthetized animal Patients receiving an overdose of chlorpropamide are known to complain of muscular weakness[10] Hypoglycemia may be a factor involved, but glucose infusions were sometimes not adequate to alleviate these symptoms On the other hand the possibility of a direct pharmacological effect of chlorpropamide on electro-



the response was s
after four hours

lyte balance muscular function, spinal reflexes, or central nervous system must be considered

The electrolyte balance was studied in dogs who received repeated sublethal doses of chlorpropamide until a state of prostration was obtained Sodium and potassium values in serum were within limits of normal with few exceptions (TABLE 2) On the other hand, it is noteworthy that these dogs appeared to respond more rapidly and more completely to combined infusions of glucose and isotonic saline solutions than to glucose alone

TABLE 2

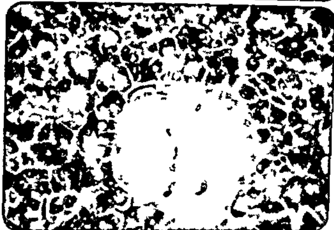
SERUM Na+ SERUM K+ (mEq /l) VALUES OF FEMALE M ... DAILY ORAL DOSES OF CHLORPR ... (Sample Taken 2 Hours Post Dose Unl ...

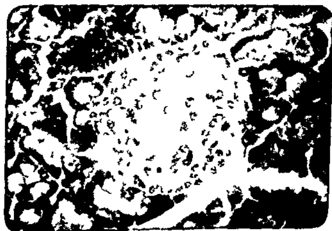
Dog No.	Daily dosage chlorpropamide mg /kg	Prechlorpropamide Na	Prechlorpropamide K+	Post Rx† Shown Na+	Post Rx† Shown K+	Post Rx Shown Na+	Post Rx Shown K+	Post R Sh Na	K			
3159	150 6× 200 16× 300 27×	156 3	5 63	Rx 5 146 2	5 28	Rx 13 147 5	5 30	Rx 21 142 4	1			
31 1	150 6× 00 16× 300 21×	not read	6 90*	Rx 5 152 0	5 83	Rx 13 144 1	5 20	Rx 145 9				
3163	150 6× 200 16× 300 12×	147 2	5 34	Rx 5 150 0	5 74	Rx 13 145 1	5 25	Rx 2 140 8	4 4	4		
3162	150 6× 200 16× 300 16×	161 4	7 78*	Rx 5 156 2	5 64	Rx 13 139 1	6 67	Rx 21 143 9	3			
2953	150 6× 200 16× 300 5× 400 1×	151 3	5 09	Rx 5 155 4	5 54	Rx 13 153 7	5 15	Rx 21 134 2	6 52	R 1		
3166	150 3× 200 10× 300 13×	154 9	5 71	Rx 4 145 1	5 00	Rx 12 140 8	5 19	Rx 19 146 5	2 94	Rx 26 150 0	4 0	r
3023	150 6× 200 16× 300 1×	146 7	5 39	Rx 5 153 4	5 25	Rx 13 148 5	5 00	Rx 21 136 7	4 58	6 hr post Rx 23 144 8	3 66	(Va u n t se r tox n ms
3173	150 4× 200 9× 300 3×	154 9	4 68	Rx 4 140 1	6 36	Rx 12 131 1	4 48	Rx 15 143 8	4 86	D ed after Rx 16 show ng severe tox c symptoms		
3160	150 6× 200 4×	138 7	5 90	Rx 5 159 3	5 05	Rx 9 140 3	6 32	D ed overn ght after Rx 10 Se ere tox c symptoms not observed				
3009	150 6× 200 4×	158 7	6 40*	Rx 5 154 0	5 58	Rx 9 97 8	5 77	Severe tox c symptoms after Rx 10				
3108	150 3× 00 4×	157 4	5 44	Rx 4 137 6	6 9	24 hr post Rx 7 145 9	5 78	(Values at onset of severe tox c symptoms)				
3000	150 3× 200 4×	149 0	5 59	Rx 4 136 13	5 86	Rx 7 141 2	4 79	Severe tox c sympt ms after Rx				

* Th se serum K+ values that are h gher than normal for the d g (3 7 to 6 mEq /l) may reflect the fact that these dogs were relat vely recent arrivals in the laboratory and the r feed ng patterns had not yet stabil zed

† Rx rep esents number of doses follow ng wh h blood was w thdrawn for analys s







In the cat nerve muscle preparation there was evidence of a direct effect of high doses of chlorpropamide on contractile force (FIGURE 10) Mono synaptic (knee jerk) and polysynaptic spinal reflexes (flexor reflex) in cats were decreased gradually however, all these effects were not sufficiently *marked to explain the degree of weakness observed in the intact animals*

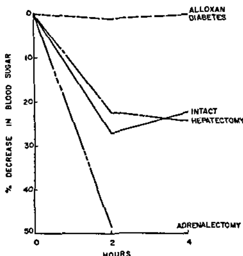


FIGURE 9 The effect of single oral doses of 100 mg /kg chlorpropamide to rats under various conditions Blood sugar determinations were made every 2 hours Fasting conditions were maintained throughout the test Six rats were used in each group and the blood sugar responses of the groups were compared at each point with the preinjection level Alloxan diabetic rats showed no hypoglycemia adrenalectomized animals responded with a large drop in blood sugar followed by death between 2 and 4 hours after drug administration No significant difference was observed in hepatectomized and intact animals

performed in cats immobilized
change in doses of as much as
by acoustic or sciatic stimulation
were not abolished Only after doses of 150 mg /kg I V or more was a slowing of the EEG noted, spindles were observed occasionally, but incon

FIGURE 8 Effect of chlorpropamide on beta cell granules of rat pancreas stained with

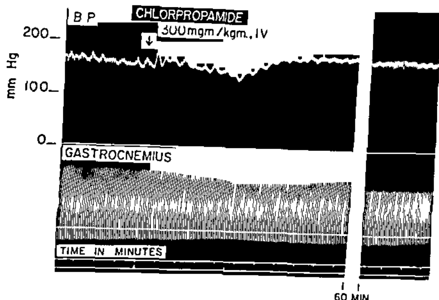


FIGURE 10 The effect of high doses of chlorpropamide on contractile force of the cat sciatic nerve gastrocnemius preparation (Nembutal anesthesia) The muscular contraction was reduced only by about 20 per cent following *intravenous administration of high* doses of chlorpropamide

sistently During these experiments, the blood pressure varied from 110 to 130 mm Hg, and showed only a slight rise after drug administration

Discussion

Although the hypoglycemic effect of chlorpropamide could be demonstrated in rats, rabbits, and dogs, the rhesus monkey was not only most sensitive to the drug but, like the human being showed a more pronounced and more prolonged hypoglycemic response than it did to tolbutamide The metabolic fate of chlorpropamide may vary from species to species and may explain these quantitative differences Such species variations in metabolism for tolbutamide are known to exist between dogs and human beings [11]

A cumulative effect may occur in dogs for the first few days following oral administration at various dose levels, but an equilibrium was reached within a few days beyond which cumulation did not progress As observed by

in the human being

From the limited data available on the fate and excretion of chlorpropamide in dogs only, few conclusions can be drawn at this time The long duration of action of chlorpropamide appears to parallel a slow rate of excretion

Chromatographic analysis of urine will determine whether chlorpropamide

is metabolized in the dog, unchanged excretion is reported for the human being [13] The difference in hypoglycemic potency between dogs, on the one hand, and human beings and monkeys, on the other, may depend upon differences in metabolism It has been suggested[4] that chlorination of the phenolic group prevented carboxylation of the compound in the para position, a reaction known to take place with tolbutamide in the human being [11]

When given chronically to rats, dogs, and monkeys chlorpropamide was tolerated without untoward side effects in doses at least twenty times the recommended human dose This represents a remarkable degree of safety for a powerful hypoglycemic agent Of special interest was the lack of histopathological changes in livers of rats and dogs treated with high doses of chlorpropamide for periods of up to twenty months These results are in agreement with findings of Root,[6] and they support the view that reversible jaundice, encountered only very rarely in human patients, is more a matter of individual idiosyncrasy than a toxic effect characteristic for chlorpropamide It may be necessary to consider such factors as history of liver disease impaired liver function prior to medication, or even increase in biliary secretion as described by Purnell *et al* [14] for the sulfonylureas

Growth inhibition as seen exclusively in the rat experiments may have various causes and may eventually offer an approach to the mode of action of chlorpropamide However, the results available at present do not allow an analysis of this drug effect Metabolic interference of chlorpropamide with cysteine and/or glycine supply to the growing organism was considered but this reaction is unlikely because addition of these amino acids to the diet did not overcome growth depression It should be remembered that nonspecific stress[15] might be a factor

[illegible]

sodium infusions than to glucose alone is yet unexplained, as no evidence of sodium deficiency could be obtained in these animals

In itself the direct effect of chlorpropamide on the contractile force of striated muscle was not definite enough to account for ataxia and weakness The same was true for the reflex depression obtained in cats

Gerstenberg *et al* [17] had observed reflex depression in rabbits with tolbutamide, an effect that seemed to be much more pronounced and was assumed to be unrelated to hypoglycemia

It is doubtful whether the EEG findings following high single doses of chlorpropamide offer an explanation for the side effects observed with toxic doses of the drug Amounts of chlorpropamide necessary to elicit these EEG changes must be considered excessive However, it is conceivable that hypoglycemia as well as central and peripheral nervous system effects

chlorpropamide are contributing to the symptoms described for the intact organism

Summary

Chlorpropamide is a potent orally active hypoglycemic agent of long duration of action Its activity may be demonstrated best in rhesus monkeys where it shows activity in the dose range used in human beings Fate studies in dogs indicate slow excretion of the compound in acute experiments

Upon chronic administration to various species of animals, chlorpropamide was found to be well tolerated in doses twenty times greater than the recommended human dose With the exception of growth retardation in very high doses in rats, the compound produced no gross anatomical or histopathological changes

Chlorpropamide is qualitatively similar to, but quantitatively more potent than, other hypoglycemic agents of the sulfonylurea types Evidence was presented that certain central nervous system effects observed experimentally after high doses, or clinically following overdosage, may be related to a direct action of the drug rather than to its hypoglycemic effect

Acknowledgments

The work reported and summarized in this paper contains data obtained from studies performed by S Y Pan, J Kaiser, M Finkelstein, K Finger, J F Snell, L Schorr, A Drakontides, and their staff, whose efforts are greatly appreciated

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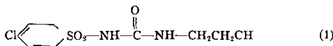
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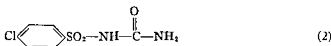
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Discussion of the Paper

Mary A Root (*The Lilly Research Laboratories Eli Lilly and Company Indianapolis Ind*) Jurg A Schneider and I are apparently in complete agreement about the pharmacology of chlorpropamide[1] However I can add a bit to his study of its metabolism All of this study of metabolism has been done by John Welles of our Toxicology Department

Schneider mentioned that paper chromatography of urine from dogs given S^{35} labeled chlorpropamide yielded three spots We have used unlabeled chlorpropamide in similar experiments and have also found three spots from dog urine The spots were eluted from the paper crystallized and then identified by x ray diffraction and infrared stud es The formulas of these three substances are


(1)


(2)


(3)

The total amount of each metabolite was determined by elution from the paper and measurement of the ultraviolet absorption on a recording spectro photometer This method is not completely quantitative but does give approximate values

In normal dogs maintained on a constant daily dose of chlorpropamide from 27 to 33 per cent of the drug (Formula 1) was excreted unchanged every 24 hours The sulfonylurea (Formula 2) accounted for from 35 to 40 per cent of the administered dose, and the sulfonamide (Formula 3) for another 16 to 24 per cent This gives a total 24 hour recovery of from 78 to 97 per cent of the daily dose These studies do not exclude the possi bility that there are other metabolites of chlorpropamide but Welles has been able to account for the major portion of the drug

The metabolic products of both carbutamide and tolbutamide have been

shown to vary greatly from species to species[2,4] For this reason we have investigated the major metabolic products of chlorpropamide in rabbits In these animals 80 to 95 per cent of a dose of chlorpropamide was excreted unchanged (Formula 1) Preliminary studies in man indicate that some of the drug is excreted unchanged but that there are probably 1 or 2 metabolic products Much more work must be done before we can establish the existence of such products and if so their identity

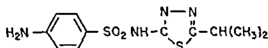
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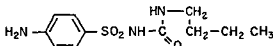
HYPOGLYCEMIC SULFONYLUREAS EFFECT OF STRUCTURE ON ACTIVITY

William M McLamore G M Fanelli S Y P an G D Laubach
Research Laboratories Chas Pfizer & Co Inc Brooklyn N Y

Although a very considerable literature has accumulated on the pharmacological and clinical properties of 2 or 3 sulfonylurea drugs little or nothing has been published until quite recently on the effects of structural changes on hypoglycemic activity within this general class [1-4] We thought it would be of interest therefore as background for this monograph to outline the results of such a study which has been under way in our laboratories for some time and which has led to the hypoglycemic agent chlorpropamide (Diabinese) Moreover although our results are in substantial agreement with those recently published there are some minor differences



2254 RP (I P T D)



CARBUTAMIDE (BZ-55)

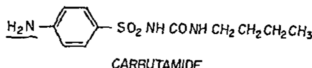
FIGURE 1

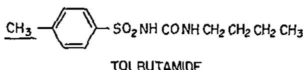
The detailed history of the hypoglycemic sulfonylureas is too well known to require repetition here One facet of this history however is pertinent to the present discussion namely the close structural resemblance (FIGURE 1) of the early sulfanilylthiadiazoles (1942)[5,6] to the later sulfonylureas (1955)[7] In view of this close similarity it is perhaps surprising that the hypoglycemic effects of sulfonylureas were discovered so much later and apparently completely by chance

Discovery of the hypoglycemic properties of 1 butyl 3 sulfanilylurea or carbutamide[7] (BZ 55 Invenol Nadisan) was soon followed by the discovery[8,9]

advantage of this structural change was the complete loss of antibacterial properties—a predictable result which was carefully *confirmed for tolbuta*

ratories and screened pharmacologically for their effect on the blood sugar





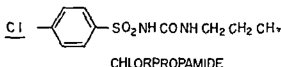


FIGURE 2

of fasting white rats at several intervals after oral administration of 100 to 300 mg /kg of the compound Our goal of course was to find a drug of superior potency and duration of action and with minimal side effects The structure activity correlations resulting from this study are outlined below

As illustrated in FIGURE 3 the generalized sulfonylurea structure can be considered in three portions (1) the alkyl chain *R* (2) the urea portion and

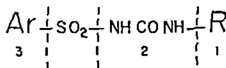


FIGURE 3

(3) the (substituted) aryl group *The effects on activity of structural changes* in each of these three portions taken one at a time are indicated in FIGURES 4 *to 8*

In most of the active series simple alkyl chains of 3 to 4 carbon atoms led to peak activity Homologues having a branched alkyl chain such as *tert* butyl or a cyclic alkyl substituent had quite good activity with a probable shift of peak activity in the cyclic case to C_5–C_7 Compounds



R	ACTIVITY
n-C_3H_7 to n-C_4H_9	+ + + to + + + +
BRANCHED C_3H_7 to C_5H_{11} (e g tert-C_4H_9)	+ + to + + + +
CYCLIC C_5H_9 to C_6H_{11}	+ + + to + + + +
NHR = $N<^{R_1}_{R_2}$, e g – N(cyclic)	+ + to + + +
R = aryl, e g –C_6H_4–Cl	+ + to + + +

Figure 4 Effect of the alkyl chain (R) on activity

CH_3–C_6H_4–SO_2–

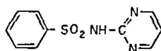
	ACTIVITY	DURATION
NH — C(=S)–NH – C_4H_9	++	+
NH — C(=O)– O – C_4H_9	+	
NH — C(=O)–CH_2 – CH_3	±	
NH — C(=O)–CH_2 – C_4H_9	±	
NH — C(=O) – C_6H_5	+	+
NH-NH C(=O)–NH – C_4H_9	±	
NH-NH-C(=O) — C_3H_7-iso	±	
N(CH_3)-C(=O)-NH – C_4H_9	±	

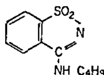
Figure 5 Effect on activity of simple changes in the urea portion

in which the N′ atom was disubstituted were moderately active. Replacement of the alkyl substituent by aryl also afforded some compounds of good activity.

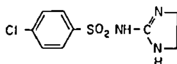
Modifications in the urea portion of the molecule that have been studied are outlined in FIGURE 5 (simple changes) and FIGURE 6 (more complex changes). Such modifications led almost uniformly to loss of activity; weak activity was retained in a few of the structural variants, notably the thiourea, the carbamate, and one of the more complex analogues.

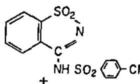
Variations in the aryl portion of the molecule (FIGURES 7 and 8) were the most fruitful of all the structural modifications studied. In general *p*-substituted aryl substituents led to compounds of maximal activity and duration of action. Unsubstituted phenyl derivatives were usually less active; *o*-substituted and di-substituted (2,4, 2,5, 3,4) phenyl compounds were still





ACTIVITY ± ±





ACTIVITY ± +

FIGURE 6. Effect on activity of more complex changes in the urea portion.

less active. Naphthylsulfonylureas were very weakly active. Compounds in which the aryl group was heterocyclic (for example, 2-thienyl, 5-chloro-2-thienyl) were, however, moderately active.

The most effective of all *p* substituents studied was halogen (Cl, Br), and especially chlorine. From a considerable number of *p*-chlorobenzenesulfonylureas with various alkyl substituents, the propyl compound was chosen on the basis of extensive animal work as the most potent and longest acting of all the sulfonylureas studied in our program. This compound has received the generic name chlorpropamide and the Pfizer trademark Diabinese.

Extensive further work in animals and in man has demonstrated this drug to be distinctly more potent and longer acting in all species than previous sulfonylureas. The relatively short duration of action of tolbutamide in man can very probably be attributed to its fairly rapid and complete metabolic

Chlorpropamide

This fact is in

)amide decrease

$Ar-SO_2NH\,CO\,NH\,R$

Ar	R	ACTIVITY	DURATION
Cl	C_3H_7 (CHLORPROPAMIDE)	++++	+++
Br	C_3H_7	++++	+
F	C_3H_7	++	++
CH_3 (TOLBUTAMIDE)	C_4H_9	+++	++
CH_3	C_4H_9	±	
CH_3, CH_3	C_3H_7	++	++
	C_3H_7	+++	++

FIGURE 7 Effect on activity of substitution in the aryl portion

$Ar-SO_2NH\,CO\,NH\,R$

Ar	R	ACTIVITY	DURATION
α, β	α C_4H_9	±	++
	β C_4H_9	+	++
S	C_4H_9	++	+
Cl, S	C_4H_9	++	+

FIGURE 8 Effect on activity of more drastic changes in the aryl portion

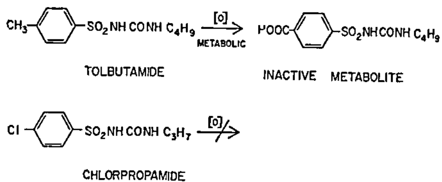


FIGURE 9

much less rapidly than those of tolbutamide and that the drug produces hypoglycemia of longer duration

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THE PHYSICAL PROPERTIES OF CHLORPROPAMIDE AND ITS DETERMINATION IN HUMAN SERUM

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Chas Pfizer & Co Inc Brooklyn N Y

Chlorpropamide [1 (*p* chlorobenzenesulfonyl) 3 propylurea Diabinese*] belongs to the class of arylsulfonylurea compounds, which have found wide spread use in the oral treatment of diabetes mellitus It was felt worthwhile to study some of the physical properties of this compound and to develop a method suitable for its determination in human serum

Chlorpropamide can be characterized by a number of physical properties The present paper deals with preliminary observations on the following (1) titration behavior, (2) melting point (3) ultraviolet absorption spectrum, (4) infrared absorption spectrum (5) solubility, (6) paper chromatography and (7) countercurrent distribution Such data proved useful in the develop ment of an assay method for this product

Several assay methods have been reported for 2 other arylsulfonylurea compounds namely, tolbutamide and carbutamide [1]† Forist *et al*,[2] Spingler and Kaiser,[3] and Bladh and Norden[4] developed methods for the determina tion of tolbutamide (1 butyl 3 *p* tolylsulfonylurea) these were based essen tially on the intense ultraviolet absorption of this compound at 228 mμ In addition, McDonald and Sawinski[5] and Spingler[6] described colorimetric procedures for the determination of tolbutamide Carbutamide (1 butyl 3 sulfanilylurea) can be determined in serum by diazotization and coupling reactions[7] The method to be described for chlorpropamide is based on the ultraviolet absorption of this compound at 232 5 mμ in 0 01 N hydrochloric acid It is isolated from serum by a two stage extraction procedure which includes (1) extraction of serum acidified with phosphoric acid with chloro form, and (2) re extraction of the chloroform phase with a dilute solution of sodium carbonate The aqueous phase from the second extraction is adjusted to *p*H 2 with hydrochloric acid the chlorpropamide content is determined by measurement of the absorbance at 232 5 mμ

Experimental

Physical Properties of Chlorpropamide

Titration Chlorpropamide is an acid, it can be titrated as such in several solvent mixtures Dioxane water, acetone water, and U S P ethanol water were found to be quite useful for this purpose, equal volumes (10 ml) of each solvent were utilized in these mixtures Chlorpropamide (70 mg) was dissolved in the dioxane water medium and was then titrated poten tiometrically with standard 0 5 N sodium hydroxide The resulting titration curve (FIGURE 1) is that of a monobasic acid A blank titration was also run

* Diabinese is the trademark of Chas Pfizer & Co Inc, for chlorpropamide

† These compounds are referred to in the literature under various designations and names (Constam[1])

on the solvent mixture alone An equivalent weight of 279 was found, that calculated for $C_{10}H_{13}N_2O_3SCl$ is 276 7 In dioxane water a $pH_{1/2}$ value of 6 5 was indicated for chlorpropamide, whereas the $pH_{1/2}$ values in acetone water and U S P ethanol water are 6 2 and 6 1, respectively The $pH_{1/2}$ value corresponds to the pH at the 50 per cent neutralization point in these titration curves These determinations were made with a Beckman Model G pH meter

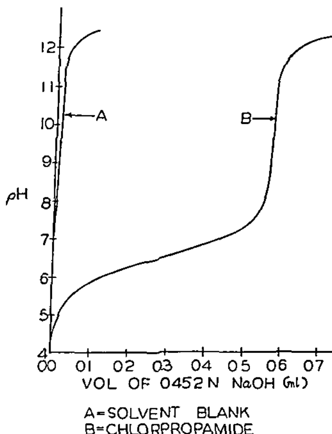


FIGURE 1 Potentiometric titration of chlorpropamide in dioxane water

Melting point Chlorpropamide melts at 127 5° to 128 5° C Previously Marshall and Sigal[8] reported a range of 127° to 129° C, and Ruschig *et al*,[9] a range of 126° to 128° C

Ultraviolet absorption spectra In 0 01 N hydrochloric acid chlorpropamide exhibits an ultraviolet absorption maximum at 232 5 mμ (ϵ = 16 5 × 10^3) and a series of less intense absorption maxima through the 250 to 280 mμ wave length region The latter occur at approximately 257 5 mμ, 265 5 mμ, 269 mμ and 277 mμ, with ϵ = 710 at 265 5 mμ (FIGURE 2) Absorbance at 232 5 mμ follows Beer's law over the concentration range 0–15 γ/ml

In U S P ethanol, chlorpropamide shows a maximum at 231 5 mμ ($\epsilon = 15\ 8 \times 10^3$) and a series of weaker absorption maxima through the 250 to 280 mμ wave length region The latter occur at approximately 258 mμ 265 mμ 268 5 mμ and 276 5 mμ with $\epsilon = 580$ at 265 mμ

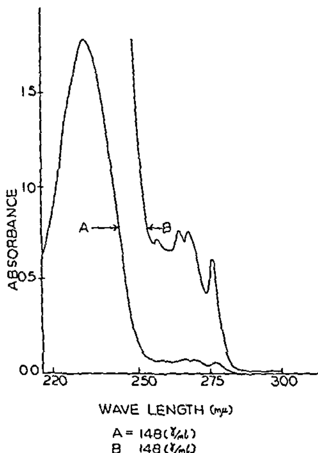


FIGURE 2 Ultraviolet absorption spectrum of chlorpropamide in 0 01 N HCl

In 0 01 N sodium hydroxide chlorpropamide exhibits an absorption maximum at 228 5 mμ ($\epsilon = 13\ 2 \times 10^3$) absorption in the 250 to 280 mμ region is not as clearly defined as in the above two solvents

Infrared absorption spectra Characteristic absorption maxima in the infrared spectrum of chlorpropamide between 2 μ to 15 μ are shown in FIGURE 3 These spectra

mull a potas

Solubility

solvents Concentrations of 25 mg/ml are readily obtainable at room

particularly in acid media The solubility of chlorpropamide in buffered (MacIlvaine) aqueous solutions at *p*H 2 2 to 6 0 was determined after 2 hours' equilibration at 25° C The amount of this compound that dissolves in this *p*H range is as follows *p*H 2 2, 0 1 to 0 2 mg/ml, *p*H 3 0, 0 1 to 0 2 mg/ml, *p*H 4 0, 0 1 to 0 2 mg/ml, *p*H 5 0, 0 3 to 0 4 mg/ml, *p*H 6 0, 2 1

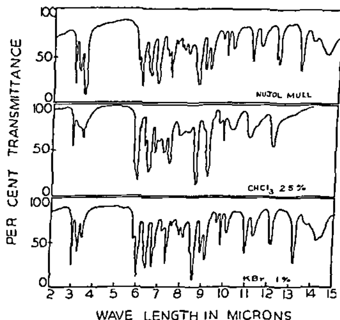


FIGURE 3 Infrared absorption spectra of chlorpropamide

to 2 3 mg/ml At *p*H values greater than 6 5 concentrations of chlorpropamide greater than 5 mg/ml are readily obtainable under these conditions

artition chromatography was used to An apparatus similar to that described re with the following solvent systems (1) mobile phase, ethyl acetate-chloroform (1 3) saturated with formamide—immobile phase, formamide methanol (1 1), Whatman No 1 paper, (2) 2 per cent K_2HPO_4 with untreated Whatman No 4 paper, and (3) mobile phase, chloroform saturated with ethylene glycol—immobile phase, ethylene glycol methanol (1 1), Whatman No 4 paper The 2 per cent K_2HPO_4 system used ascending solvent flow Solutions of chlorpropamide were spotted on paper in amounts containing 50 to 100 γ of compound, at times as much as 1000 γ was streaked on the paper After development and drying, spots

were detected either by their ultraviolet absorption through a zinc silicate-coated fluorescent screen[11] or by spraying with blue tetrazolium reagent [12] After spraying with the blue tetrazolium reagent, the sheet is warmed a short time in an oven (90° C), chlorpropamide exhibits a whitish zone on a general blue background Single zones were observed in these systems In the

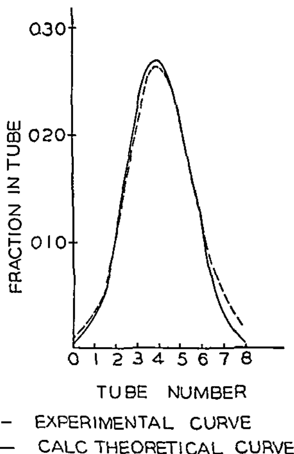


FIGURE 4 Countercurrent distribution of chlorpropamide between chloroform and *p*H 6 7 buffer

ethyl acetate chloroform (1 3) system the R_f value was approximately 0 1 in the 2 per cent K_2HPO_4 system, approximately 0 8 0 9, and in the chloroform ethylene glycol system, approximately 0 2

Countercurrent distribution The homogeneity of chlorpropamide was tested by means of the technique of countercurrent distribution This compound was distributed between chloroform and Clark and Lubs *p*H 6 7

buffer Under these conditions the distribution coefficient of chlorpropamide is about 1 An 8 plate transfer was run, using a chloroform solution of chlorpropamide (concentration of 300 γ/ml) At the end of the distribution the chlorpropamide content of each phase in each tube was determined through ultraviolet spectrophotometry When the fraction of the total chlorpropamide content per tube was plotted against the corresponding tube number, a typical distribution curve was obtained in good agreement with a calculated curve (FIGURE 4)

Microanalytical data Chlorpropamide can be recrystallized readily from acetone water mixtures Typical microanalytical data are calculated for $C_{10}H_{13}N_2O_3ClS$ C, 43 40, H, 4 74, N, 10 12, S, 11 59, Cl, 12 81 Found C, 43 48, 43 45, H, 4 75, 4 84, N, 10 17, 10 02, S, 11 99, 11 84, Cl, 12 6

Determination of Chlorpropamide in Human Serum

Reagents Five reagents were used in this study

(1) 0 067 M Phosphoric acid 4 55 ml of concentrated phosphoric acid (Mallinckrodt, A R, 85 per cent) diluted to a total volume of 1000 ml with water

(2) Chloroform Mallinckrodt A R

(3) One per cent sodium carbonate Mallinckrodt, A R, 10 gm of anhydrous sodium carbonate per 1000 ml of solution

(4) Hydrochloric acid (a) 0 4 N (b) 0 01 N

(5) Chlorpropamide stock solution 200 mg of chlorpropamide are slurried with 30 ml of distilled water and sufficient 0 1 N NaOH is added (with stirring) until a *p*H of 7 is reached The resulting solution is transferred quantitatively to a 100-ml volumetric flask and diluted to the mark with distilled water It is stable for at least one week when stored at room temperature This stock solution serves as a convenient starting point in the preparation of solutions for use in recovery experiments and the determination of the $E^{1\%}_{1cm}$ value of chlorpropamide

Apparatus In this experiment the following apparatuses were used (1) centrifuge tubes of 10- and 40-ml capacity, glass-stoppered and graduated, (2) a centrifuge capable of handling the centrifuge tubes, and (3) a spectrophotometer, either the Beckman Model DU spectrophotometer or the Cary Model 11 recording spectrophotometer quartz cells (1 or 2 ... I for absorbance measurements

It should be men...
out this assay ... chloroform
and dried prior

Procedure O ... of serum is mixed with 2 ml of 0 067 M phosphoric acid and 4 ml of water in a 40-ml centrifuge tube The resulting acidified serum is then extracted with 10 ml of chloroform by gentle shaking for about 15 seconds * The phases are separated by centrifugation, and 5 ml of the

* Occasionally an emulsion forms at this stage which is not easily broken by centrifugation If this occurs an additional 10 ml of chloroform are added to the centrifuge tube and the contents are reshaken and centrifuged Instead of a 5 ml aliquot a 10-ml aliquot of the clear chloroform layer is taken and the experiment continued as above

chloroform layer is transferred to a 10-ml centrifuge tube containing 5 ml of 1 per cent sodium carbonate. The contents of this centrifuge tube are shaken and then centrifuged. A 4 ml aliquot of the sodium carbonate layer is transferred to a 10-ml volumetric flask, neutralized with 2 ml of 0.4 N hydrochloric acid and diluted to the mark with 0.01 N HCl. The absorbance value of this solution is determined at 232.5 mμ using either the Beckman DU spectrophotometer or the Cary spectrophotometer. The reference solvent is 0.01 N hydrochloric acid. As a blank, a serum sample (devoid of chlorpropamide) is processed through this same procedure and measured in the same fashion. If a Cary recording spectrophotometer is available, it is well occasionally to record the ultraviolet spectra of the sample and blank over the 220 mμ to 300 mμ wave length interval.

Calculation. Chlorpropamide concentration

$$(\gamma/\text{ml}) = \frac{(A_S - A_B)_{232.5\,m\mu}(10^6)}{(l)(E^{1\%}_{1\,cm})(4)}$$

where l is the cell path in centimeters, A_S and A_B are the absorbance values of serum sample and blank, and $E^{1\%}_{1\,cm}$ is the absorbance of a solution of chlorpropamide in 0.01 N hydrochloric acid at a concentration of 1 gm per 100 ml when measured in a 1 cm cell at 232.5 mμ.

Recovery experiment. Known amounts of chlorpropamide (50 to 250 γ/ml) were added to human serum and then were processed through the above assay procedure. The results of a typical recovery experiment are shown in TABLE 1. Here recovery is essentially quantitative. Experiments on a

TABLE 1
RECOVERY OF CHLORPROPAMIDE ADDED TO HUMAN SERUM

Chlorpropamide added (γ/ml)	Chlorpropamide found (γ/ml)	Recovery (percentages)
250	248	99
250	248	99
250	248	99
125	127	102
125	123	98
125	123	98
50	51	102
50	50	100
50	51	102

variety of serum samples to which chlorpropamide had been added in varying amounts (37 to 564 γ/ml) yielded a recovery of 99 ± 1.4 per cent.* It is recommended that rec[illegible]
familiarize one with the
to determine the $E^{1\%}_{1\,cm}$
tory conditions.

* Average ± average deviation.

Serum blanks. A serum sample obtained from the same subject before the administration of chlorpropamide when processed through the normal procedure serves as a convenient serum blank. A variety of serum samples (free of chlorpropamide) from both diabetic and nondiabetic subjects was carried through the above extraction procedure and gave apparent chlorpropamide concentrations of 13 ± 2.7 γ/ml.

Reproducibility experiment. Serum samples from several patients being treated with chlorpropamide were combined. Ten individual determinations were made on this pooled serum sample. These results are summarized in TABLE 2.

TABLE 2
REPRODUCIBILITY OF ASSAY ON COMBINED SERUM SAMPLES

Assay No.	Chlorpropamide content (γ/ml)
1	191
2	194
3	191
4	193
5	194
6	191
7	198
8	191
9	195
10	194
	193 ± 1.8

Identification of Chlorpropamide in Human Serum

In order to demonstrate the presence of chlorpropamide in human serum the following isolation experiment was conducted. Serum samples from 16 diabetic subjects were pooled to provide 30 ml. of serum with a chlorpropamide content of 138 γ/ml. This pooled serum sample was acidified to pH 2 to 3 with 0.067 M phosphoric acid and was extracted with chloroform [illegible]

now shaken with a volume of 1 per cent sodium carbonate such that a fourfold increase in concentration was effected. This 1 per cent sodium carbonate layer was removed, neutralized with 0.4 N HCl, and diluted so as to provide two solutions comparable in concentrations with those shown in FIGURE 2. The ultraviolet absorption spectra of these 2 solutions were recorded on a Cary Model 11 spectrophotometer and were found to be identical with those of chlorpropamide. After the determination of the ultraviolet absorption spectrum, these solutions were combined and extracted with chloroform. The chloroform extracts were concentrated to dryness in a stream of nitrogen and the residue was dried at reduced pressure over potassium hydroxide

The infrared spectrum of this residue was determined in chloroform in a microcell on a Ba rd recording spectrometer and it was identical with that of chlorpropamide Furthermore the paper chromatographic behavior of this residue was the same as that of chlorpropamide in the 2 per cent K HPO_4 system

Interferences The method described for the determination of chlor propamide is not specific for this compound In order to test for possible interferences the following compounds were added to 1 ml of serum in the quantities indicated and were carried through the usual extraction procedure
(1) *p* chlorobenzen
sulfonamide (0 25
(5) β hydroxybuty
(7) salicylic acid (0 5 mg) The ultraviolet absorption spectra of the resulting solutions were examined in the wave length interval 220 to 350 mμ using the Cary Model 11 recording spectrophotometer In the case of sodium *p* chlorobenzenesulfonate acetone and ethyl acetoacetate the resulting spectra were comparable to that of a serum blank However the ultraviolet absorption spectrum of the solution obtained from the β hydroxybutyric acid experiment exhibited strong end absorption thus making it difficult accurately to determine chlorpropamide in the presence of 5 5 mg /ml of this compound With *p* chlorobenzenesulfonamide acetylsalicylic acid and salicylic acid major absorption maxima were found at 228 mμ 227 mμ and 235 5 mμ respectively indicating that these compounds would present a serious interference in the procedure used to determine chloropropamide

Summary

Chlorpropamide is an acidic substance as demonstrated by potentiometric titration Its ultraviolet absorption spectrum shows a maximum at 232 5 mμ in 0 01 N hydrochloric acid with less intense maxima in the 250 to 280 mμ wave length region The infrared absorption spectrum of chlorpropamide is characterized by a number of strong absorption bands through the 2 to 15 μ wave length region It is soluble in a variety of common organic solvents but exhibits a limited solubility in water at *p*H values below 5 Its homogeneity can be demonstrated by paper partition chromatography and counter current distribution

A method is presented for the determination of chlorpropamide in human serum It consists of a two stage extraction procedure that includes an extraction of acidified serum with chloroform followed by re extraction of the chloroform phase with dilute sodium carbonate After suitable acidification of the sodium carbonate phase the chlorpropamide content is determined by measurement of the absorbance at 232 5 mμ Recoveries of chlorpropamide from human serum and serum blank values are quite acceptable The manipulations involved in the assay are relatively simple and determinations can be carried out in reasonably short time intervals Chlor

β hydroxybutyric acid p chlorobenzenesulfonamide acetylsalicylic acid and salicylic acid would present interferences

ACKNOWLEDGMENTS

We acknowledge our sincere appreciation to T M Lees G W Walsh F R LaPietra R J Hickey W H Boegemann and H J Pazdera of the Chas Pfizer & Co Inc staff for their contributions to the work described in this paper

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METABOLIC FATE OF CHLORPROPAMIDE IN MAN*

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We have investigated the urinary excretion, blood levels protein binding, and chromatographic behavior of chlorpropamide, a new sulfonylurea This was accomplished by administering this drug to nondiabetics and to diabetic patients Work by others has shown that tolbutamide another sulfonylurea has a serum half time of 4 hours and that it is carboxylated at the *p* methyl group *in vivo*[1,2] On the other hand carbutamide has a longer serum half time of 33 hours and the *p* amino group is acetylated *in vivo*[3] To our knowledge there are no published reports concerning the metabolic fate of chlorpropamide in man

Methods

The radioactive chlorpropamide used in these experiments had an initial specific activity of 66 6 mg mc It was given as a water solution with sufficient carrier added so that 20 μc of sulfur 35 equaled 250 mg of chlorpropamide This solution was given to patients as follows

Group 1 Four nondiabetic patients without renal disease were given a single oral dose of 250 mg of radioactive chlorpropamide containing 20 μc of sulfur 35

Group 2 Three stable diabetic patients subject No 1 45 years old received 500 mg tolbutamide twice daily subjects Nos 2 and 3 71 and 64 years old respectively received 250 mg of chlorpropamide daily as therapy On the day of the test each patient received orally one dose of 250 mg of radioactive chlorpropamide No further chlorpropamide was given during the test period

Group 3 (*a*) A 41 year old diabetic patient who had not received chlorpropamide prior to this test was given intravenously 20 μc of radioactive chlorpropamide without added carrier One tablet of 250 mg of chlorpropamide was given orally at the same time (*b*) A 71 year old diabetic was treated in the same way as the patients of Group 1

The radioactive samples were counted with an automatic micromil gas flow G M tube with an average background of 14 cpm using a preset count of 1000 Sufficient recounts were obtained to give a 5 per cent average counting error or in the case of the weaker samples a sufficient number of recounts to obtain at least 3 count rates that were within 1 cpm of the median count rate The mean of these median values was considered to be the true count rate Each sample was weighed and corrected for self absorption All serum samples were deproteinized by sodium tungstate and sulfuric acid precipitation prior to plating

* The work reported in this paper was supported in part by a grant from Chas Pfizer & Co Inc Brooklyn N Y

The binding of chlorpropamide as determ
200 µg of chlorpropar
serum These aliquo
37° C for 24 hours
at *p*H 7 6 for 48 hours The per cent of the radioactivity remaining at the end of the dialysis was considered as the percentage of bound chlorpropamide The binding of 4 types of protein solutions was determined as follows (1) normal serum, (2) commercial serum albumin in 2 concentrations, 25 and 50 mg /ml , and (3) serum obtained from a patient with marked hypoalbuminemia His serum protein was 4 7 gm per cent Electrophoretic analysis showed that 2 6 per cent of this protein was albumin, 6 5 per cent alpha 1 globulin, 17 per cent alpha 2 globulin, 23 per cent beta globulin, and 51 per cent gamma globulin *In vivo* binding was determined by giving a diabetic patient 250 mg of radioactive chlorpropamide orally Serum samples were obtained at 4 and 48 hours These samples were dialyzed in the same way as the protein solutions

The chromatographic method used for the detection of chlorpropamide in blood and urine was a modification of the method of Seligson and Shapiro for α keto acids[3] The only significant change was elimination of the incubation with 2-4 dinitrophenylhydrazine A chlorpropamide standard was used to locate the chlorpropamide spots under ultraviolet light after chromatography Numerous unidentified spots seen under ultraviolet light after chromatography of extracts of urine are unrelated to the administration of chlorpropamide

To determine whether erythrocytes absorbed chlorpropamide, we incubated 25-ml samples of fresh whole blood at 37° C for 2 hours in siliconized vessels gassed with 5 per cent CO_2 and 95 per cent O The plasma was separated by centrifugation The erythrocytes were resuspended and washed in cold normal saline 4 times prior to the determination of radioactivity

The serum concentration of chlorpropamide was measured by the technique of Toolan and Wagner[4] This method makes use of a chloroform extraction of acidified serum with 0 067 M phosphoric acid The chloroform extract is then washed with aqueous 1 per cent sodium carbonate The aqueous sodium carbonate is acidified to *p*H 2 with HCl, and the optical density is measured at 232 5 mµ The optical density of the fasting serum is subtracted and the net optical density is compared to a standard The average optical density of the serum of 5 normals who had not received this drug previously gave an apparent chlorpropamide value of 17 µg /ml by this method

Results

TABLE 1 contains the serum radioactivity values obtained from 4 nondiabetic patients following the ingestion of 250 mg of chlorpropamide containing radioactive sulfur 35 Maximum serum radioactivity and optical
The log of the
nects the mean
The first and

more rapid of these has an uncorrected half time of 32 hours, the slower component has a half time of approximately 16 days However, since only 2 data points were obtained, this value is only a rough estimate If the effects of the slower component are removed from the initial slope, a half time of approximately 24 hours is obtained These data show that the serum

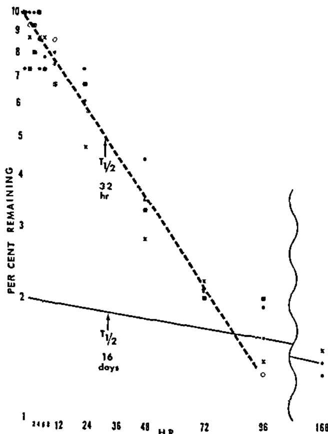


FIGURE 1 Disappearance of serum radioactivity from the serum of 4 nondiabetic patients In this figure, 10 is equal to 100 per cent

disappearance of chlorpropamide occurs at 2 separate rates. Half times

rst 3 subjects
The fourth

TABLE 1
SERUM RADIOACTIVITY CORRECTED TO PEAK ACTIVITY
Disappearance of radioactive chlorpropamide from the serum of 4 nondiabetic patients The maximum serum radioactivity has been made equal to 100 The actual maximum serum radioactivity is listed at the bottom of the table

Hours	Subject No 1	2	3	4	Mean
1	100	100	73	100	
2	87	93	100	73	
4	93	93	100	80	
6	87	86	73	100	86
8	87	73	73		78
12	67	86	80	67	75
24	47	57	73	67	61
48	28	36	44	33	35
72	21	21	21	20	21
96	14	13	19	20	16
168	15		13		14
Net cpm/ml serum at 100%	90	84	90	81	

TABLE 2
SERUM RADIOACTIVITY CORRECTED TO PEAK ACTIVITY
[illegible] The
n [illegible]

Subject No	Group No 2: 1	2	3	Group No 3: 1	2
Hours					
1	100	55	92	100	82
2	85	100	100	76	100
3				92	
4	85	64	83		88
5				76	
6	100	64	75		82
8	85	73	83	72	70
24	77	54	50	64	59
48	69	45	42	40	41
72	46	36	25	28	35
96	54	36	25	20	35
Net cpm/ml serum equal to 100%	60	66	72	150	72

subject received this drug intravenously and the fifth subject received this drug in the same way as the patients of Group 1 the data from this table are shown in FIGURE 2 It can be seen that the early disappearance of the radioactivity from the serum in all but one subject was similar to that of the nondiabetic patients given this drug However all values fall outside of 2

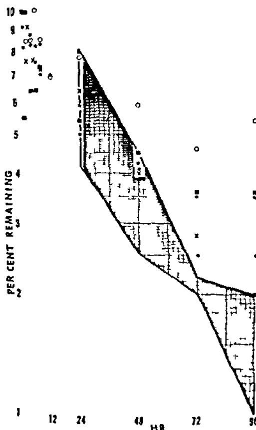


FIGURE 2 Disappearance from the serum of radioactive chlorpropamide in 5 diabetic subjects In this figure 10 is equal to 100 per cent

standard deviations from the mean of the normal group after the 48-hour specimen Whether this higher percentage of slowly excreted chlorpropamide represents differences between the handling of the drug in normals and diabetics cannot be determined from these data since the groups are not comparable It is notable that the disappearance of the radioactivity from the serum of subject No 1 was much slower than that of the other subjects This diabetic patient preloaded with tolbutamide had a normal phenol sulfonphthalein excretion urinalysis and blood urea nitrogen His serum

uric acid level was elevated but there was no other evidence of gout except that he had had an episode of pain in the right ankle of several days' duration on one occasion 2 months prior to these studies

TABLE 3 presents the percentage of the ingested dose excreted in the urine of the nondiabetic subjects in Group 1 The patients are presented in the same order as they were in TABLE 1 so that a comparison can be made between

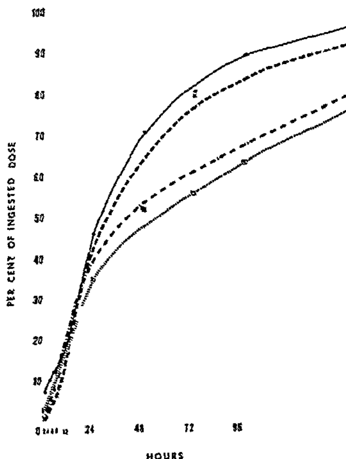


FIGURE 3 Cumulative urinary excretion of radioactive chlorpropamide in 4 nondiabetic patients

the serum and urinary radioactivity FIGURE 3 shows these data plotted linearly The mean cumulative excretion during the 96 hours of the tests was 77 per cent This agrees well with the ser... values which showed 16 per cent of the radioac... ...um 96 hours after the ... ...n the patients without ... the urine ... the urinary excretion of the radioactivity

TABLE 3

CUMULATIVE URINARY EXCRETION OF RADIOACTIVE CHLORPROPAMIDE BY FOUR NONDIABETIC SUBJECTS

The values presented are the percentages of the ingested dose

Subject No	1	2	3	4
Hours				
2	0 7	1 3	7 1	2 6
4	2 7	3 6	10	5 2
6	4 9	8 4	12	7 8
8	6 4	11	13	12
12	17	21	19	17
24	42	41	46	35
48	60	54	71	52
72	80	62	81	56
96	84	68	90	64

TABLE 4

CUMULATIVE URINARY EXCRETION OF RADIOACTIVE CHLORPROPAMIDE BY FIVE DIABETIC SUBJECTS

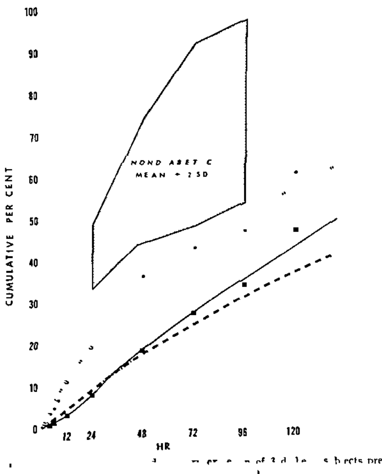
The results are presented as percentages of the ingested dose

	Group No 2			Group No 3	
Subject No	1	2	3	1	2
Hours					
2	0 4		1 2	3 0	3 9
4	1 7	0 5	3 4	8 2	5 1
6	3 0	1 3	4 7		13
8	3 7		6 8	12	15
12	4 5	3 0	11	17	22
24	8 5	7 8	19	36	35
48	17	18	36	66	55
72	24	27	43	88	66
96	31	34	47	92	75
120		47	61		79

obtained from the diabetic patients of Groups 2 and 3 Their serum values are presented in the same order as in TABLE 2 In FIGURE 4 the urinary excretion of these preloaded patients was markedly less than that of the normal group However, it should be noted that the 2 groups are not completely comparable in regard to certain other factors such as age and the presence of diabetes Consequently, the differences in the preloaded group and the group not loaded prior to testing could be due to other factors The mean cumulative urinary excretion of the nondiabetics of Group 1, plus and minus 2 standard deviations is presented on this figure for comparison

FIGURE 5 shows the cumulative urinary excretion of the other 2 diabetic

patients of Group 3 The patient with the highest urinary excretion received this drug as therapy up to the day of the test On the day of the test he received an intravenous tracer dose of chlorpropamide without added carrier followed by a 0 25 gm tablet of chlorpropamide His rapid excretion of the drug is markedly different from that of the other 3 diabetics The other excretory curve was obtained from a patient given 1 oral dose of this drug



[illegible] of 3 d[illegible] s[illegible]bjects pre[illegible]

His excretory pattern is very similar to that obtained from the 4 patients in the *normal* group

Fecal excretion of an intravenously injected chlorpropamide was determined in 1 diabetic patient who was already taking this drug as therapy Carmine red dye given at the same time as the tracer dose appeared in his 48-hour stool specimen The patient s fecal excretion of chlorpropamide was 0 52 per cent on day 1 0 92 per cent on day 2 and 0 74 per cent on day 3

It would appear that fecal excretion is not a very significant excretory pathway.

TABLE 5 shows the results of our dialysis experiments. *In vitro* binding by a normal subject was 6.9 per cent. A patient with a total serum protein of 4.7 gm per cent with almost no albumin showed binding of 3.1 per cent which is less than one half of that of the normal serum. Albumin solutions

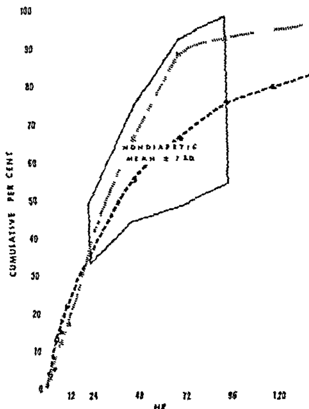


FIGURE 5. Cumulative chlorpropamide urinary excretion of 2 diabetic subjects. The mean ± 2 S.D. of the mean of the normal group is presented for comparison.

of 2.5 and 5.0 gm per cent bound 2.2 and 4.6 per cent, respectively. These data suggest that both albumin and globulin participate in this binding. However, no complete serum fractionation studies have been done. We did not find binding of chlorpropamide by crystalline insulin solutions of 1 to 4 mg/ml (22 units/mg). These concentrations of insulin are considerably less than those of the other protein solutions; therefore binding of a degree equal to that of albumin could have been missed. Nondialyzable chlorpropamide was present in the serum of a diabetic patient who had ingested this drug. These values approximate those of the *in vitro* experiments.

TABLE 5
PROTEIN BINDING OF RADIOACTIVE CHLORPROPAMIDE MEASURED BY DIALYSIS
Determined by dialyzing serum protein samples at *p*H 7 6 for 48 hours

	Per cent bound
	(Mean ± standard error)
In vitro	
Normal	6 87 ± 0 66
Hypoalbuminemic	3 08 ± 0 15
Albumin 2 5 gm %	2 23 ± 0 08
Albumin 5 0 gm %	4 60 ± 0 33
In vivo	
Diabetic 4 hours	4 35 ± 0 60
Diabetic 48 hours	6 96 ± 0 99

TABLE 6
EFFECT OF INCREASING CONCENTRATIONS OF CHLORPROPAMIDE ON PROTEIN BINDING
Each sample was incubated for 24 hours at 37° C and dialyzed at *p*H 7 6 for 48 hours

Concentration (µg /ml)	Normal serum		Serum albumin (50 mg /ml)	
	Per cent	µg	Per cent	µg
25	7 8	1 9	6 1	1 5
50	6 6	3 3	6 6	3 3
100	4 9	4 9	4 9	4 9
200	13	26	11	22

In an attempt to determine the effect of varying concentrations of chlorpropamide on these binding phenomena, we added 25 to 200 µg of chlorprop- [illegible] TABLE 6 sho s that varying the concentrat [illegible]

With the chromatography system used by us the sulfur-35 labeled chlorpropamide standard exhibited 3 separate radioactive areas Little difference was noted in the R_f values of the standard and that obtained from serum and urine specimens The R_f values obtained are shown in TABLE 7 Seventy to 85 per cent of the radioactivity was associated with the R_f value 0 9 in the standard and in the extracts of urine and serum This area of radioactivity corresponds to the major area of ultraviolet light absorption

The incubation of chlorpropamide with blood resulted in a plasma radio-

TABLE 7

CHROMATOGRAPHY OF SULFUR-35 LABELED CHLORPROPAMIDE

Chromatography of serum and urine specimens following the ingestion of chlorpropamide Peak No 1 contained 70 to 85 per cent of the radioactivity Peak No 2 contained 10 to 25 per cent of the radioactivity, and Peak No 3 contained 0 to 5 per cent of the radioactivity

Peak No	1	2	3
Chlorpropamide standard	0 9	0 4	0 2
2 Hour serum	0 8	0 5	
8 Hour serum	0 9	0 3	
2 Hour urine	0 9	0 4	0 2
8 Hour urine	0 9	0 4	

activity of 15 730 net counts per minute (ncpm) and an erythrocyte radioactivity of 70 ncpm After 2 hours of incubation the plasma count was 16 120 ncpm, and the erythrocyte count was 100 ncpm This slight difference in radioactivity in the erythrocytes could be accounted for easily by experimental variations in the washing of the erythrocytes In confirmation of these data, erythrocytes obtained from a patient given an oral dose of radioactive chlorpropamide failed to show absorbed radioactivity

Discussion

The disappearance of radioactive chlorpropamide from the sera of the 4 nondiabetic patients in Group 1 showed at least 2 separate disappearance rates These dual disappearance phenomena exhibited by chlorpropamide

not been reported to occur following the ingestion of carbutamide and tolbutamide

Extrapolation of the slower component back to zero time suggests that approximately 20 per cent of chlorpropamide could be present as this slowly excreted component, this is 3 times greater than values obtained in the binding experiments Whether this slower component merely represents chlorpropamide bound to serum proteins or metabolized chlorpropamide cannot be determined from these data Our evidence would suggest that this slowly

ve

on

he

1

shows that significant differences in the time required to excrete this drug exist among diabetic patients receiving this drug as therapy

The urinary excretion of radioactive chlorpropamide closely parallels its disappearance from the serum The excretion of chlorpropamide by the preloaded diabetic patients was not as uniform as that of the nondiabetic patients This would suggest that significant differences exist in the excre

tion of this drug by various diabetic patients Because of the possible cl importance of these differences and because this may be related to the dis state further study should be made of this phenomenon Unlike car mide or tolbutamide this compound does not seem to be metabolized pr excretion since no definite chromatographic differences were noted a serum urine and standard

Binding of carbutamide and tolbutamide by serum proteins has not reported Chlorpropamide appears to be bound to both serum albumi globulin This binding is related to the concentration of chlorprop: present in the serum and approximately equal binding can be pro *in vitro* The presence of binding by proteins may account for the odd tion space determinations obtainable after ingestion of this drug By mining the space of distribution of intravenously administered chlor amide in two 70 kg diabetic patients 10 and 11 l spaces were obt: These values do *not correspond to either the extracellular or intravas* space and correcting it for amount of chlorpropamide bound by the s proteins still fails to produce a space of distribution comparable to reported for carbutamide or tolbutamide We can surmise that bindi proteins other than serum proteins or leakage into the cells must occur failure to find absorption of chlorpropamide by *in vitro* incubation of ery cytes does not rule out concentration of this drug by other tissues

Summary

(1) We have used chlorpropamide labeled with sulfur 35 to determine metabolic fate of this drug

(2) The disappearance of chlorpropamide has an initial apparent half of 32 hours for the major portion of the dose and a half time of 16 days for remainder Chlorpropamide is excreted almost entirely by the kidn Only very slight fecal excretion was demonstrated

(3) Chlorpropamide is made partially nondialyzable by serum prote Increasing the concentrations of chlorpropamide in the serum increases binding

(4) We could not detect metabolic alteration of chlorpropamide prior excretion

Acknowledgments

The technical assistance of Thomas Redding Ronald Lee and Ber Wulff and the art work of Shirley Shipman are greatly appreciated

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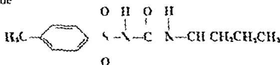
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Discussion of the Paper

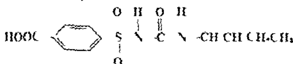
STEFAN S FAJANS (*University of Michigan, Ann Arbor Mich*) In view of the almost complete excretion of chlorpropamide in urine reported by Philip Johnson I shall mention briefly some published and unpublished observations about the metabolism and excretion of tolbutamide

In 1956 we reported upon the findings of a major urinary excretion product of tolbutamide after its administration to normal and diabetic subjects Upon acidification of the urine of such subjects to a *p*H 5 or less a white precipitate appeared This was shown to be the carboxylic acid derivative of tolbutamide (1 butyl 3 *p* carboxyphenyl sulfonylurea) as reported by us in conjunction with Struck and others of the Upjohn Research Laboratories Kalamazoo Mich

Tolbutamide



Tolbutamide excretion product



On very large dosages of tolbutamide such as 6 gm per day a maximum of about 75 per cent of the administered drug could be recovered from the urine as this metabolic oxidation product This was determined by acidifying the urine and then drying and weighing the purified insoluble precipitate

In order to rule out additional urinary metabolites of tolbutamide and to determine more quantitatively the urinary excretion of the carboxyl compound a joint project was set up with W L Miller of the Upjohn laboratories at his suggestion It was planned to give S^{35} tolbutamide to normal and diabetic subjects and to examine the urine for metabolites of tolbutamide by paper chromatography

As can be seen from TABLE 1 3 gm of unlabeled tolbutamide was given daily to 6 subjects for 5 days and 15 mg of S^{35} tolbutamide was administered on the fourth day Urines were collected for 24 hours on the fourth and fifth days The chromatographic studies were performed by W L Miller

The results of these studies are given in the table It is apparent from *the data for the first 4 subjects (3 control subjects and 1 tolbutamide non* responsive diabetic subject) that 90 per cent or more of the administered drug was excreted as the carboxyl sulfonylurea within the first 24 hours and nearly 100 per cent within 48 hours Furthermore the carboxyl excretion product was the only human transformation product of tolbutamide identified

The lesser amounts of radioactivity recovered in the urine of the last two diabetic patients could well be accounted for by unfortunate errors in urine collection on the hospital wards. In these two patients there was also no evidence of excretion of a metabolite other than the carboxyl substituted tolbutamide.

In summary, it can be stated that

(1) the carboxylic acid derivative of tolbutamide is the only urinary excretion product in man, and (2) on the basis of data from the first four subjects, it appears that tolbutamide is quantitatively excreted in the urine as this metabolite.

TABLE 1
URINARY EXCRETION OF S^{35} CARBOXYL SULFONYLUREA AFTER ORAL ADMINISTRATION OF S^{35} TOLBUTAMIDE*

Subjects	S^{35} Excretion, per cent		
	0 to 24 hours	24 to 48 hours	0 to 48 hours
Normal control			
T D	99 0	1 0	100 0
G R	95 6	4 3	99 9
J D	89 8	10 0	99 8
Tolbutamide nonresponsive diabetic			
R J	90 0	9 0	99 0
Tolbutamide responsive diabetics			
R C	—	—	90 6
N M	73 1	6 9	80 0

* Tolbutamide was administered 3 gm per day for 5 days S^{35} tolbutamide, 15 mg on fourth day. Urine collections were made on fourth day (0 to 24 hours) and fifth day (24 to 48 hours)

RELATIVE POTENCIES OF CHLORPROPAMIDE AND TOLBUTAMIDE IN MAN

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In a previous publication[1] we have reported the results of experiments in which the hypoglycemic potencies of chlorpropamide and tolbutamide in 10 normal subjects were compared at varying intervals after administration of the drugs by mouth. One gram of chlorpropamide produced responses that exceeded those to 1 gm. of tolbutamide at 1, 2, 3, 4, 6, 12, and 18 hours after administration. More recently the venous blood glucose levels[2] of 20 normal subjects were determined 1 hour after each subject had received 1 gm. of tolbutamide on one occasion, and after 1 gm. of chlorpropamide on another occasion at intervals of 1 week. The order in which the drugs were administered was alternated. The drugs were administered orally in tablet form together with 4 ounces of water. The mean difference between the control fasting blood glucose values and the blood glucose levels 1 hour after tolbutamide averaged 6.6 mg. per cent, while chlorpropamide produced responses averaging 14.1 mg. per cent. This difference was highly significant ($p < .001$). Since it seems unlikely that either drug had been appreciably metabolized or excreted as early as 1 hour after administration by mouth, the greater potency of chlorpropamide is probably not due entirely to its slower rate of degradation and excretion. The fact that chlorpropamide was definitely more potent 1 hour after administration suggests the possibility that it is more potent than tolbutamide even when serum levels are comparable. However, the levels of the drugs in the serum were not determined in this experiment. Consequently, it is possible that the greater potency of chlorpropamide is partially due to the fact that it is more readily or more completely absorbed.

FIGURE 1 shows the responses of 6 normal subjects to varying doses of chlorpropamide and tolbutamide when the blood glucose levels were measured 2 hours after the doses were administered in random order at intervals of not less than 4 days (those subjects who received 2 gm. of chlorpropamide were not given another dose for at least 1 week). Each of the subjects received 4 different doses of each drug, with the exception that only 3 of the subjects received a dose of 2 gm. of chlorpropamide. In each instance the fasting blood glucose was compared to the blood glucose 2 hours after a drug.

It may be noted that, with each of the doses, the responses were greater to chlorpropamide than to tolbutamide. Tolbutamide, 0.25 gm., produced no response, while 0.5 gm. of the drug produced responses comparable to those after 0.25 gm. of chlorpropamide. Tolbutamide, 1 gm., and chlorpropamide, 0.5 gm., also produced similar responses. It is of interest that tolbutamide, 2 gm., produced responses that were not greater than those to 1 gm., which suggests the possibility that the point of maximum response was being

reached. However, the data are not adequate to establish this point. On the other hand, the responses to the various doses of chlorpropamide were roughly related to the log of the dose.

Since the biological half time of chlorpropamide is several times longer than that of tolbutamide (Johnson *et al*, in this monograph), it seems likely that differences in the responses to chlorpropamide and tolbutamide would be far greater than those demonstrated here if the effects were measured 12 to 48 hours after comparable doses of the 2 drugs.

That the high serum levels present for several days after a single dose of chlorpropamide are associated with proportionate physiological activity of

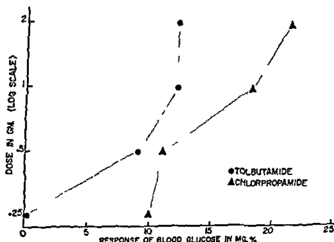


FIGURE 1. Hypoglycemic potencies of tolbutamide and chlorpropamide in normal subjects. Each plot represents the mean response of 6 subjects 2 hours after the oral administration of the drug except that only 3 of the subjects received 2 gm. of chlorpropamide. The patients who received chlorpropamide also received tolbutamide. See text for details.

the drug is suggested by the following example. We administered chlorpropamide, 2 gm., to one of our normal subjects, whose blood glucose was measured 1 hour later and proved to be 44 mg. per cent. At 2 hours it was

not be entirely aroused from sleep. The state of semiconsciousness persisted for 1 hour before being relieved immediately by the intravenous injection of 50 gm. of glucose. The level of serum chlorpropamide at this time was 16.4 mg. per cent.

Serum Levels of Orally Administered Chlorpropamide

(Johnson *et al*, in this

some way partially bound to a substance or substances in the plasma Elsewhere in this volume Johnson *et al* indicate that chlorpropamide is to some extent bound to protein If this is the case it is probably not appropriate to refer to a chlorpropamide space If however it were assumed that the drug was homogeneously distributed in a space the data in TABLE 1 would indicate that such a space would necessarily be less than 9 1 in volume since there is at least 115 mg in every liter of plasma 2 hours after oral administration of 1 gm of the drug

The Effect of Chlorpropamide on Glucose Tolerance

One of the strongest arguments against the direct or indirect peripheral action of the sulfonylurea agents has been the failure to demonstrate an effect of single doses of tolbutamide on glucose tolerance [4, 5] On the other hand TABLE 2 shows that chlorpropamide produced in each of 5 subjects a statistically significant increase in the rate of disappearance of intravenously administered glucose Venous blood glucose determinations were carried out 10 [illegible] the test was repeated in the same manner except that chlorpropamide 0 5 gm was injected with the glucose The control mean disappearance rate was 1 22 per cent per minute After chlorpropamide the mean disappearance rate was 1 85 per cent per minute One of these subjects weighing 178 pounds had a control rate of 1 6 per cent per minute which increased 2 4 per cent per minute after chlorpropamide 0 5 gm It is of interest that the

TABLE 1
SERUM LEVELS OF CHLORPROPAMIDE AFTER 1 GM ORALLY

Subject	Weight (lbs)	Serum chlorpropamide	
		1 Hour (mg %)	2 Hours (mg %)
1	155	3 6	13 0
2	145	5 8	10 9
3	124	2 9	10 6
4	1 8	6 9	12 0
5	185	8 3	12 7
6	114	7 0	10 3
7	145	8 2	10 7
Mean	149	6 1	11 5

intravenous administration to this subject of 1 8 units of glucagon free insulin produced a strikingly similar change (2 5 per cent per minute)

Discussion

It is not possible to establish accurately the ratio of the potencies of chlor propamide and tolbutamide from the data in FIGURE 1 In the first place, too few subjects have been tested to establish precisely a dose response curve for either drug However it is clear that in a certain range of dosage chlor propamide produced an effect approximately comparable to that produced by doses of tolbutamide twice as large As mentioned previously, it may be assumed that the difference in the responses to chlorpropamide and tolbutamide on a milligram for milligram basis could be expected to be much greater if the tests were done from 12 to 48 hours after administration of the drugs because of the longer biological half time of chlorpropamide (Johnson *et al*, in this monograph) The difference in potency might be expected to be par ticularly marked if the response to the drugs were compared after daily administration in which case there would be a cumulative effect for a few days with chlorpropamide and no appreciable cumulative effect with tolbutamide

It should be stressed that the subjects who received chlorpropamide (FIGURE 1) were the same who received tolbutamide This paired-control design will, of course, make it possible to identify significant differences in the dose response curve with a fewer number of tests since the factor of the vary ing responsiveness from subject to subject is eliminated At present we are doing a paired-control study in which responses of normal subject to 0 25 gm chlorpropamide are being compared to their responses to 0 5, 1, and 2 gm of tolbutamide This experiment will continue until statistically significant differences are identified

The degree to which studies of potencies of sulfonylurea derivatives in nor mal subjects are applicable to the establishment of their potencies in various

TABLE 2
THE EFFECT OF 0 5 GM CHLORPROPAMIDE ON INTRAVENOUS GLUCOSE TOLERANCE

Fasting blood glucose (mg)		10 min after glucose* (mg)		40 min after glucose (mg)		Glucose disappearance rate % per minute†	
Control	Chlorprop	Control	Chlorprop	Control	Chlorprop	Control	Chlorprop
70	70	163	173	135	134	0 57	0 75
70	67	148	130	100	159	1 08	1 82
2	59	158	146	88	25	1 48	2 76
66	63	130	130	82	65	1 23	1 67
3	71	146	130	69	42	1 76	2 25
Mean 70	66	149	142	95	65	1 22	1 85
						P < 0 04	

* Fifteen gm injected in 1 to 2 min timed from start of injection
† Assuming that the per cent per minute disappearance during a 30-min period occurred at a constant rate

types of diabetic subjects is not known. In general, however, we have found in studies with carbutamide, tolbutamide, chlorpropamide, and metahexamide that there has been a good correlation between the potencies of the drugs in normal subjects and their potencies in diabetics of the maturity-onset type. However, this matter has not been systematically evaluated by us, and the opinion expressed here is based only on our "clinical impressions" and requires verification by more direct studies.

Summary and Conclusions

Chlorpropamide, 1 gm, produced significantly greater hypoglycemic responses in normal subjects 1 hour after ingestion than did tolbutamide, 1 gm. Chlorpropamide 0.25 gm and 0.5 gm, produced hypoglycemic responses in normal subjects 2 hours after ingestion comparable to doses of tolbutamide twice as great. Since it is unlikely that either drug had been appreciably metabolized or excreted 1 to 2 hours after ingestion, the relatively greater potency of chlorpropamide probably is not attributable entirely to its longer biological half time.

Seven normal subjects had serum chlorpropamide levels averaging 11.5 mg per cent 2 hours after ingesting chlorpropamide, 1 gm, indicating that about one third of this dose was present in the plasma at that time.

Chlorpropamide 0.5 gm significantly increased the rate of disappearance of intravenously injected glucose. In one subject the effect of an intravenous injection of chlorpropamide 0.5 gm, on the glucose disappearance rate was strikingly similar to that of 1.8 units of insulin administered intravenously.

Two findings that have resulted from studies carried out since this report

tolbutamide were administered intravenously the hypoglycemic effects during the first hour were not significantly different.

Acknowledgments

The authors are indebted to B. J. Matter, Charles Key, Dorothy A. Wood and Robert Henry for technical assistance in carrying out these studies.

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EFFECTS OF ADMINISTRATION OF CHLORPROPAMIDE UPON THE CARBOHYDRATE METABOLISM OF ISOLATED TISSUES*

D W Clarke

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It has been shown previously that the administration of carbutamide to rats over a 14-day period can cause a slight increase in the level of glycogen in the diaphragm [1] If this diaphragm is removed and incubated in a suitable medium there is a decrease in the glycogen level, and this decrease is greater in diaphragms from treated animals than in those from control animals This larger drop is probably only a reflection of the higher initial level of glycogen, but the higher initial level is compatible with the theory that carbutamide acts by increasing the effective insulin level in the plasma, possibly by increasing insulin secretion by the pancreas A similar amount of tolbutamide does not cause a statistically significant increase, although the experimental data suggest such an effect.

This type of experiment has been repeated using chlorpropamide, to learn whether similar increases in muscle glycogen could be obtained The drug was made up as either a 2½ per cent or a 1¼ per cent suspension in 1 per cent methyl cellulose (Methocel) solution and was force fed daily to rats in measured amounts so that a dose of either 250 mg/kg/day, or 125 mg kg day was given At the same time, Orinase† and RP 2259‡ were given to other rats in a dosage of 250 mg/kg/day Control rats received only the Methocel They were kept on this regime for from 5 to 7 days and were then sacrificed

Results

The rats used in these tests were divided into a group of adult rats (Group 1) and a group of younger rats that were still growing (Group 2) The average weight changes are shown in the first two lines of TABLE 1 Generally speaking the effect of the hypoglycemic agents is to cause a weight loss in the older animals or a decreased weight gain in the younger ones

The results of measurements on initial glycogen levels in the diaphragm, on glycogen decrease in the diaphragm after incubation in a saline phosphate medium on glucose uptake of the diaphragm, on liver glycogen and glucose release by a liver slice on plasma potassium and on plasma glucose are further shown in TABLE 1 Results from the 2 groups of animals are pooled, since there seemed to be no difference between the younger and the older animals

An analysis of the variance shows that there is a significant difference

* The work reported in this paper was supported in part by grants from Chas Pfizer & Co Inc Brooklyn N Y and from Poulenc Ltd Montreal, Canada

† Tolbutamide

‡ 2 Sulfamido p aminobenzene 5 tert butyl 1 thio 3 4-diazole

between the values for initial glycogen Chlorpropamide (at its high level) and RP 2259 seemed to cause a small increase Neither Orinase nor chlorpropamide (at its low dose level) show such an effect

In a second experiment chlorpropamide was given 350 mg /kg day for 14 days If anything there was a drop in diaphragm glycogen level 2 99 mg gm for controls 2 77 mg /gm for treated animals The average weight gain for treated animals was 34 gm , compared to an average gain of 83 gm for control animals The test animals were in a poorer nutritional state and this may account for their failure to show an increase in muscle glycogen

An additional experiment in which Orinase and RP 2259 were given,

value of 2 04 mg gm which is not significantly greater than the control level

TABLE 1

	Control	RP 2259	Orinase	Chlorpropamide		No of animals
				1¼%	2½%	
Weight changes (grams)						
Group 1	6 3	−6 3	−2 7	−8 0	−7 7	3
Group 2	25 0	25 3	12 0	19 7	21 7	3
Initial diaphragm glycogen (mg gm)	2 75	3 65	2 57	2 57	3 27	5
Glycogen decrease from original level (mg gm)	1 30	1 20	1 02	0 96	1 49	5
Liver glycogen (mg gm)	9 8	8 6	9 4	9 3	12 4	5
Liver glucose production (mg gm)	11 9	10 5	9 2	10 3	8 3	5
Glucose uptake (mg /gm)	1 64	2 01	1 56	1 53	1 32	3
Blood glucose (mg %)	150	150	135	143	125 (162)	6
Plasma potassium (mEq ml)	6 18	6 11	6 37	6 45	5 99	4

The values for the first experiment (TABLE 1) show that there were no significant changes in glycogen losses in the diaphragm following incubation in glucose uptake by the diaphragm in liver glycogen or glucose production by ood sugar values show tha The value shown for the value omitted The average value with this peculiar high value included is shown in brackets The experiments with the higher doses of the hypoglycemic agents showed that chlorpropamide caused a drop in liver glycogen to 57 per cent of the

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[illegible]

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An additional experiment in which Orinase and RP 2259 were given 400 mg kg day showed that after 3 days neither agent had any effect upon

value of 2 04 mg /gm which is not significantly greater than the control level

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control value Orinase caused a drop to 42 per cent of the control value, where RP 2259 caused a drop to 60 per cent of the control value

In Vitro Studies

The influence of chlorpropamide *in vitro* on certain tissues was also studied The glucose output of liver slices from normally fed rats was measured when this compound was added to the incubation medium in different concentration Slices were incubated in a Dubnoff shaker for 45 min at 37° C in 10 ml of phosphate saline buffer The results are shown in TABLE 2

TABLE 2

	Chlorpropamide concentration mg /ml			
	0 00	0 06	0 25	1 00
Glucose output mg gm	9 45	8 91	9 65	9 92
Per cent change from control		−5 7	+2 1	+5 0

The inhibitory action of chlorpropamide on glucose output is evident at a concentration of the drug that is about the same as that reported in the blood of human subjects This suggests that chlorpropamide may act at least in part, by inhibition of liver activity No further studies were done to elucidate the reason for the increase in glucose output with a higher concentration of chlorpropamide

Chlorpropamide can also inhibit the oxygen uptake of certain tissue Kidney slices from normal rats were incubated in a Krebs-Ringer phosphate medium in a Warburg apparatus, and oxygen utilization was measured After 1 hour chlorpropamide was added, and it was found that at a concentration of 0 1 mg /ml it gave a 19 per cent inhibition in the rate of oxygen uptake at 0 01 mg /ml it gave a 14 per cent inhibition

When added to a rat liver homogenate at a concentration of 10^{-3} M (0 28 mg /ml) in the presence of cytochrome *c* and succinate[2] there was a drop of 7 per cent in the oxygen consumption rate, at 10^{-2} M (2 8 mg /ml) a drop of 13 per cent was observed

Conclusion

Chlorpropamide may, under certain conditions slightly increase the glycogen content of the rat diaphragm Under similar conditions Orinase has no effect RP 2259 has a greater effect Any stimulating action upon glycogen synthesis is consistent with the hypothesis that certain oral hypoglycemic agents act by increasing the plasma insulin activity

Chlorpropamide in low concentration can cause a small reduction in the glucose output of liver slices and a reduction in oxygen utilization by kidney slices and liver homogenates It is suggested that part of the hypoglycemic action of chlorpropamide may come about through its hepatic action

Acknowledgments

We are indebted to Chas Pfizer & Co, Inc, for supplies of chlorpropamide and to Poulenc, Ltd for supplies of RP 2259

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IN VIVO EFFECT OF INSULIN AND CHLORPROPAMIDE ON CERTAIN LIVER ENZYMES IN RATS

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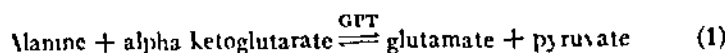
In the over all animal economy insulin is known to be protein sparing a property that has not as yet been associated with the newer oral hypoglycemic agents The mechanisms by which insulin exerts its effects on protein metabolism are still unclear but it has been assumed that these effects are secondary manifestations of the alterations in carbohydrate metabolism known to occur in hypoinsulinism and hyperinsulinism The present study was prompted by several reports[1, 2] wherein it was proposed that the gluco neogenic action of the adrenal corticosteroids is based on their ability to stimulate the hepatic activity of glutamic pyruvic transaminase (GPT) one of the two major transaminase systems involved in protein metabolism The effect of insulin and chlorpropamide on GPT and glutamic-oxaloacetic transaminase (GOT) activity seemed worthy of study The results presented below compare the effects of these two hypoglycemic agents on GPT and GOT activity with those of hydrocortisone Lactic dehydrogenase (LDH) activity was also studied as an example of an enzyme system not directly concerned with protein metabolism

Experimental

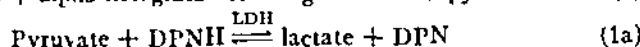
Eighty two albino rats weighing from 100 to 200 gm were used for this study Food and water were offered freely unless otherwise indicated Protamine zinc insulin and chlorpropamide (sodium salt) were administered in varying doses of from 1 to 4 U kg and from 50 to 100 mg /kg , respectively Hydrocortisone acetate was administered in a dosage of 5 mg per animal With the exception of one preliminary acute experiment, all drugs were given subcutaneously in 7 daily doses over an 8 day period

On the day of sacrifice all food and drugs were withheld The animals were stunned and exsanguinated The livers were removed immediately and portions were weighed for dry weight and enzyme assays Those portions removed for assay were immediately frozen in a dry ice alcohol bath and stored at −20° C prior to analysis A 5 per cent homogenate of the sample for assay was prepared in the cold and suitable dilutions for each assay were prepared

GPT activity was measured by a modification of the method of Wroblewski[3] In this determination pyruvic acid formed in REACTION 1 is reduced to lactate after the addition of excess LDH and in the presence of reduced diphospho pyridine nucleotide (DPNH)

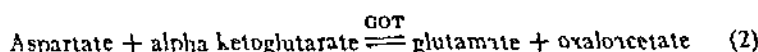


$$\text{Alanine} + \text{alpha ketoglutarate} \overset{\text{GPT}}{\rightleftharpoons} \text{glutamate} + \text{pyruvate} \quad (1)$$

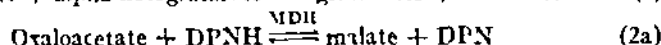


$$\text{Pyruvate} + \text{DPNH} \overset{\text{LDH}}{\rightleftharpoons} \text{lactate} + \text{DPN} \quad (1a)$$

GOT activity was measured by a modification of Karmen's procedure[4] In this assay oxaloacetate formed by REACTION 2 is reduced to malate by an excess of malic dehydrogenase (MDH) and in the presence of DPNH as in REACTION 2a



$$\text{Aspartate} + \text{alpha ketoglutarate} \overset{\text{GOT}}{\rightleftharpoons} \text{glutamate} + \text{oxaloacetate} \quad (2)$$



$$\text{Oxaloacetate} + \text{DPNH} \overset{\text{MDH}}{\rightleftharpoons} \text{malate} + \text{DPN} \quad (2a)$$

LDH activity was determined by a modification of a procedure based on REACTION 1a[5] The oxidation of DPNH in each of these reactions was followed spectrophotometrically at 340 mμ One unit of activity for all enzymes represents a change in optical density of 0 001 O D units/min Although all reactions were carried out within a fairly narrow temperature range, the results were corrected to a temperature of 25° C by a separately determined series of temperature correction factors, a procedure found desirable in previous studies[6,7]

Aliquots of the original homogenates were analyzed for nitrogen by the micro Kjeldahl method Although not always presented, results were calculated in terms of wet weight dry weight, and nitrogen content

Results

Preliminary experiments were performed to ascertain the acute effect of the administration of insulin and chlorpropamide on liver enzyme activity Three pairs of rats were fasted for 18 hours before use Crystalline insulin (2 U /kg) was adm [illegible]

[illegible]

served as a control No significant differences in hepatic enzyme activity were found between the control and treated animals (TABLE 1) despite markedly depressed terminal blood sugars in the latter groups

The absence of any change in this acute experiment is in line with what has been found in studies concerning the acute effects of insulin administration[8] Hastings *et al*[9] have concluded that insulin exerts immediate metabolic effects only at the periphery, and that its effect on hepatic metabolism is delayed and gradual Further experiments reported herein were all extended to eight days

Effects of insulin, chlorpropamide, and hydrocortisone in rats fed ad libitum TABLE 2 presents the results of one experiment evaluating the effect of insulin (1 U /kg /day) and chlorpropamide (50 mg /kg /day) given in 7 injections over an 8 day period The actual results for each animal are illustrated here to demonstrate the variability of individual measurements and to compare the results on the basis of wet weight, dry weight, or nitrogen

TABLE 1
EFFECT OF ACUTE ADMINISTRATION OF INSULIN AND CHLORPROPAMIDE ON HEPATIC ENZYME ACTIVITY

Treatment	Enzyme activity U/mg dry wt		
	GPT	GOT	LDH
(a) No treatment	84.4	973	1420
(b) Insulin (regular), 4 U/kg	83.5	1000	1450
(c) Chlorpropamide (Na), 100 mg/kg	78.8	910	1315

TABLE 2
VARIABILITY IN HEPATIC ENZYME ACTIVITY IN RATS

Treatment	Enzyme activity (units)								
	GPT			GOT			LDH		
	1*	2*	3*	1	2	3	1	2	3
(a) Control	19.3	64.7	609	213	715	6720	442	1480	13,900
	21.5	72.3	658	203	633	6230	438	1470	13,400
	23.1	74.5	663	237	764	6790	403	1300	11,600
Mean	21.3	70.5	645	218	704	6580	428	1417	13,000
(b) Insulin (PZI) 1 U/kg	12.5	39.5	359	211	666	6063	342	1080	9,827
	15.6	46.3	414	206	612	5464	399	1180	10,600
	15.6	48.1	405	207	648	5370	381	1170	9,900
Mean	14.6	44.6	393	208	642	5630	374	1140	10,100
(c) Chlorpropamide 100 mg/kg	19.3	58.8	553	197	601	5640	305	930	8,740
	16.9	54.0	469	199	636	5530	356	1140	9,900
	15.8	51.3	451	152	494	4350	419	1360	12,000
Mean	17.3	54.7	491	183	577	5180	360	1140	10,200

* Columns 1, 2, and 3 represent units/mg wet weight, dry weight, and nitrogen, respectively

content Final evaluation of the effects of therapy were made on the combined data from this and subsequent experiments on 27 animals

The results, shown in TABLE 3a indicate that hydrocortisone has a differing action on the 2 hepatic transaminases studied Thus, although steroid administration apparently had no effect on GOT activity, it resulted in a marked and significant ($p < 0.01$) increase in GPT activity These latter

TABLE 3

EFFECT OF THERAPY ON HEPATIC ENZYME ACTIVITY IN FED AND RESTRICTED ANIMALS

Treatment	Enzyme activity (± S.E.) U/mg dry wt		
	GPT	GOT	LDH
(a) Ad libitum feeding			
Control	67.4 (±1.9)	701 (±10)	1300 (±25)
Chlorpropamide 50-100 mg/kg	55.8 (±1.8)	565 (±5.3)*	1090 (±13)
Insulin (PZI) 1-4 U/kg	56.2 (+1.7)	572 (±7.1)*	1200 (±16)
Hydrocortisone 5 mg	181 (+8.6)*	689 (+36)	1430 (±88)
(b) Ad libitum feeding plus hydrocortisone			
Hydrocortisone alone	181 (±8.6)*	689 (+36)	1430 (±88)
Hydrocortisone plus chlorpropamide 100 mg/kg	214 (+4.2)*	584 (±12)*	1200 (+21)
Hydrocortisone plus insulin (PZI) 2 U/kg	195 (±7.6)*	524 (+33)*	1170 (±29)
(c) Restricted feeding			
No treatment	125 (±2.8)*	976 (±22)	966 (±14)*
Hydrocortisone	197 (+3.7)*	755 (±15)*	1090 (±29)
Chlorpropamide 100 mg/kg	101 (±2.8)*	857 (+22)	1130 (±37)
Insulin (PZI) 2 U/kg	110 (±4.3)*	923 (+31)	984 (±35)*

* Significantly different from control values ($p < 0.01$)

findings are in accord with numerous other reports on the effect of adrenocorticosteroids on the glutamic pyruvic transaminase[1,2,10,11] Although GOT activity has been reported to be unaffected by similar therapy[1,11,12] in line with our present findings there have been conflicting reports[2,13] This disagreement however may be a matter of degree, since it is the consensus that GOT activity is much less markedly influenced by steroids if at all than is GPT

In contrast to these findings the administration of insulin (1 to 4 U/kg/day) and chlorpropamide (50 to 100 mg/kg/day) were both shown to be accompanied by a significant decrease ($p < 0.01$) in hepatic GOT activity as shown in TABLE 3*a* GPT activity although apparently somewhat depressed was not significantly affected ($p > 0.18$) No reports of similar *in vivo* studies on the effects of insulin or chlorpropamide on hepatic transaminases have come to our attention However our observations would

seem to be in accord with the report by Copenhaver *et al*[14] that hepatic GOT activity is markedly elevated in alloxan diabetic rats

d

chlorpropamide on GOT activity was due to a direct hormonal influence on the liver or an indirect type of response due to changes in the periphery Hydrocortisone together with either insulin or chlorpropamide was administered to a group of 17 animals to compare the effects of a combination of

day) resulted in changes in enzyme activity that reflected the influence of the steroid as well as the hypoglycemic agents Thus GPT activity was increased as a result of hydrocortisone therapy despite the simultaneous administration of the hypoglycemic agents As shown in TABLE 3a GOT activity was depressed after the injection of insulin or chlorpropamide despite the administration of hydrocortisone Lactic dehydrogenase activity

Starvation
nt the hepatic
activity of both glutamic oxaloacetic and pyruvic transaminase[15] The effect of hydrocortisone insulin and chlorpropamide on this increase in enzyme activity was studied in a group of thirty two animals

TABLE 3c shows that even the limited food restriction imposed in the present experiments was capable of enhancing the activity of the two transaminases under consideration The administration of hydrocortisone to these animals resulted in a further augmentation of GPT activity beyond that produced by food limitation alone GOT activity on the other hand was apparently decreased to those levels encountered in ad libitum fed animals These findings further substantiate previous indications that hydrocortisone has a diverse effect on these two closely related enzymes Contrary to the findings with animals on a free diet neither insulin nor chlorpropamide could be shown to exert any influence on the activity of these enz

1 unaffected
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however resulted in a significant decrease ($p < 0.01$) in the level of this enzyme in contrast to the increases observed in the level of both transaminases It is interesting that limitation of dietary intake was the only

experimental procedure capable of altering the activity of the lactic dehydrogenase enzyme

Discussion

The results obtained in this investigation must be interpreted with caution. Much additional work is needed to elucidate the role that these enzymes play in whole body homeostasis. One fact stands out, namely, that the action of chlorpropamide paralleled that of insulin in all experiments. The cause and physiological significance of these changes in hepatic enzymes, however, are more difficult to evaluate.

The early work of Bach and Holmes[15] showed that the addition of insulin to liver slices of normal fasted rats inhibited the transformation of certain amino acids into carbohydrates. Jensen and Gray,[16] employing both *in vitro* and *in vivo* techniques, found that insulin exerts an inhibitory effect on hepatic amino acid dehydrogenase activity in normal rats. In their *in vivo* experiments insulin administration led to a 25 per cent decrease and adrenal cortical extracts to a 205 per cent increase in the activity of this enzyme. Since Stadie *et al*[17] showed that the enzyme influenced by insulin affects only the unnatural D isomer of amino acids, the physiological importance of these findings is doubtful. However, it is interesting to note that a similar increase in GPT and a decrease in GOT activity were obtained in this study after hydrocortisone and insulin, respectively. This similarity would appear to throw some doubt on the view that the change in transaminase activity after steroid therapy is a reflection of specific hormonal control.

This latter concept was suggested by Rosen *et al*[1] as an explanation of the marked increase in GPT activity and subsequent gluconeogenesis accompanying the administration of cortisone. The increase in enzyme activity after food restriction might be construed as a reflection of a stress induced oversecretion from the adrenal cortex. The differences in the action of insulin and chlorpropamide under the conditions of restricted and unrestricted feeding make this interpretation unlikely.

Starvation, however, like hydrocortisone therapy, is accompanied by an alteration in the metabolism of proteins toward catabolism. Insulin appears to stimulate anabolism. It has already been suggested that hepatic transaminase activity is inversely related to protein synthesis,[10, 18] a view that appears to accord with current findings. Similarly, Recant and Fischer[9] recently presented data indicating that tolbutamide, as well as insulin, stimulates the incorporation of tagged glycine into the liver proteins of starved animals. These [illegible]

changes in enzyme activity are under direct hormonal control or whether they represent [illegible]

tion[20] [illegible]

processe[illegible]

the findings of Zuchlewski and Gaebler[21] who demonstrated that the transfer of isotopic nitrogen from alanine to other amino acids occurred to the same

extent in the livers of normal and hypophysectomized rats treated with growth hormone despite a marked reduction in the GPT activity of the hormone treated animals Similar studies on the effects of hydrocortisone insulin and chlorpropamide might yield a more definitive concept of the mechanisms by which these play a role in protein metabolism and its relation to gluconeogenesis

Summary

Insulin and chlorpropamide act similarly in moderately depressing glutamic oxaloacetic transaminase activity in the livers of well fed animals The simultaneous administration of hydrocortisone appears to have no effect on this action of insulin and chlorpropamide

Glutamic pyruvic transaminase activity augmented by the administration of hydrocortisone alone continued to be markedly increased despite the simultaneous administration of insulin and chlorpropamide

Food restriction increased the activity of both transaminases Under these conditions neither insulin nor chlorpropamide could be shown to exert any effect on GOT activity despite the further augmentation of GPT activity by hydrocortisone

Further study is necessary to determine whether these changes in transaminase activity induced by these drugs are secondary in nature or whether they represent direct hormonal control

Acknowledgment

We acknowledge the valuable technical assistance we received from Carmine J Caruso in the experiments reported in this paper

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SOME EFFECTS OF TOLBUTAMIDE AND CHLORPROPAMIDE *IN VITRO**

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Introduction

Although a detailed discussion of available information concerning the site of action of the h...

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consider well documented the position that the acute administration of the arylsulfonylureas leads to a decrease in the pancreatic insulin content[1] and to an increase in the insulinlike activity of pancreatic vein blood[2-4] Increased circulating insulinlike activity, however, has not as yet been established in man[5,6] Whether the same sequence of events holds true during long-term, chronic administration of these compounds is as yet unknown, as is the relative physiological importance of the postulated increased insulin secretion—either acutely or during long term administration We also consider as well establ shed the ... that the h...

mildly stated—disputed

The group for which I am speaking, associated with George W Thorn's Metabolic Unit at Harvard Medical School and the Peter Bent Brigham Hospital, was impressed early by the existence of significant differences in the metabolic states induced by insulin and the sulfonylureas, respectively, even when the severity of the induced hypoglycemia was of similar magnitude and achieved in a similar period of time[5,7] If one tends to accept the insulin stimulation theory of sulfonylurea action as the major site of action of these substances, this paradox may be resolved by endowing endogenous insulin secreted into the portal vein with special, previously unknown properties[8] If, on the other hand, one is not particularly impressed by the evidence gathered for the over all physiological importance of the insulin stimulation theory of sulfonylurea action, one is led logically to give prominence to the extrapancreatic effects of the compounds Our group still favors the latter interpretation, but I stress the fact that the following observations are not presented as additional evidence for or against the physiological importance of sulfonylurea induced insulin secretion Since heated discussion on this

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point seems rather fruitless at present, it is hoped that the effects shown will contribute to the over all pharmacodynamic description of these interesting agents Indeed it may be permissible to expect that careful analysis of the extrapancreatic effects of the sulfonylureas, whatever their physiological significance may lead as well to the description of a mechanism for the sulfonylurea induced release of insulin [9]

Material and Methods

We are here concerned exclusively with the effects of tolbutamide and chlorpropamide on ketogenesis by liver slices from rats fasted 48 hours, and with the drugs effects on glucose metabolism by adipose tissue from fed normal rats The methodology employed has been described previously in some detail [10, 11] Alloxan diabetes was induced by the intravenous injection of 40 mg kg alloxan following a 48 hour fast Random blood glucose values exceeding 350 mg per cent within 2 or more weeks following alloxan injection were considered as indicative of the presence of diabetes Total pancreatectomies were performed according to Scow [12] and only those animals exhibiting both hyperglycemia and ketosis 36 hours following pancreatectomy were used

Results

Buris Boshell of our laboratory has reported[10] that tolbutamide in concentrations of 1 0 mg /ml and 0 2 mg ml significantly inhibits the formation of ketones by rat liver slices *in vitro* The previously reported data are shown in TABLE 1 It was considered of particular significance that this

TABLE 1*
EFFECT OF TOLBUTAMIDE UPON KETOGENESIS BY LIVER SLICES FROM NORMAL RATS FASTED 48 HOURS

Addition to medium	No of observations	Ketogenesis *in vitro*	
		Micrograms acetone equivalent gm wet liver 90 min	Control (percentages)
No addition	7	610 ± 63†	100
Insulin 0 1 U /ml	7	590	97 ± 4†
Tolbutamide 1 0 mg /ml	7	226	38 ± 4
No addition	8	539 ± 100	100
Tolbutamide 0 2 mg /ml	8	376	70 ± 6

* Data from Boshell et al [10] Incubations were carried out in Krebs bicarbonate buffer modified by replacing some Na^+ by K^+ to final concentrations of $Na^+ = 83$ and $K^+ = 60$ ml 1 1

† Mean ± standard error of the mean Since all experiments were *paired* experiments, the significance of the tolbutamide effect is calculated on the basis of the figures representing percentage of the control value

tolbutamide effect could be demonstrated at a concentration (0 2 mg /ml or 20 mg per cent) quite frequently encountered in plasma or serum of tolbutamide treated patients and that the add ...
centrations was ineffectiv
tation of insulin mediatio

These observations have since been extended to include a greater range of concentrations and the comparison of chlorpropamide effects with the effects of tolbutamide As shown in FIGURE 1, tolbutamide and chlorpropamide were approximately equally effective in depressing *in vitro* ketogenesis by liver slices from rats fasted 48 hours, although the data do not exclude

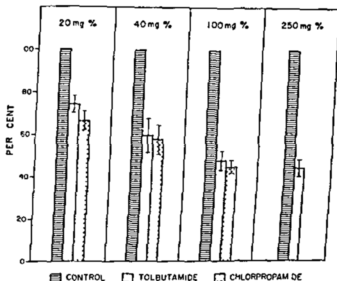


FIGURE 1 Effect of tolbutamide and chlorpropamide on ketogenesis by liver slices from rats fasted 48 hours Top figures refer to the concentrations of drugs used expressed as milligrams per 100 ml Incubations were carried out in a Krebs bicarbonate buffer modified by adjusting the concentrations of K^+ to 60 and Na^+ to 83 mEq /l

the possibility that a differential potency of the two substances might have been detected at lower concentrations (for example, 10 mg per cent) The absence of a striking potency difference with regard to ketogenesis *in vitro* thus tentatively supports the view that *in vivo* potency differences of the 2 compounds are more likely to be related to differences in the metabolic fate of the drugs rather than to differences in their intrinsic metabolic activities

FIGURE 2 further demonstrates the effects of tolbutamide and chlorpropamide upon hepatic ketogenesis as a function of time It is evident that the effect appears early and persists throughout the period of incubation The figure also demonstrates that the over-all drug induced inhibition is really greater than that previously estimated, since a major portion of the ketones present in the incubation medium (at the end of the incubation in the presence of tolbutamide and chlorpropamide) represents ketones

Whereas correlation between hypoglycemic activity *in vivo* and antiketogenetic activity *in vitro* is at least consistent with the possibility of a common mechanism of action, further studies carried out in liver slices obtained from animals in which experimental diabetes had been induced by alloxan injection or by pancreatectomy failed to support this conclusion As shown in FIGURE 3, tolbutamide effectively suppressed hepatic ketogenesis in the

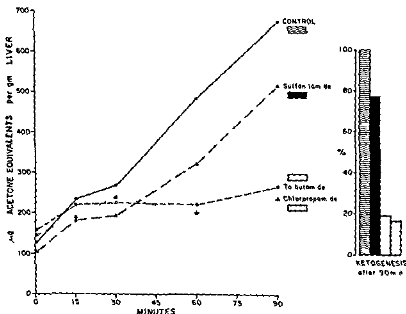


FIGURE 2 Effect of tolbutamide (100 mg per cent) chlorpropamide (100 mg per cent), and sulfanilamide (100 mg per cent) on ketogenesis by liver slices from rats fasted 48 hours Each curve represents the mean of 2 experiments

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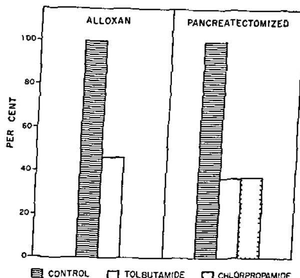


FIGURE 3 Effect of tolbutamide and chlorpropamide (100 mg per cent) on ketogenesis by liver slices from alloxan diabetic and pancreatectomized rats Shown are the means of four (alloxan) and six (pancreatectomized) experiments

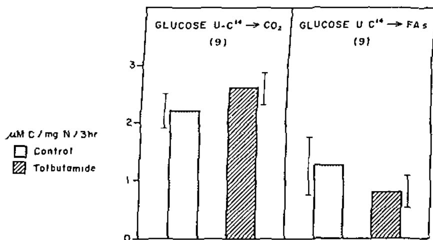


FIGURE 4 Effects of tolbutamide (0 8 mg /ml) upon rat adipose tissue *in vitro* The standard errors shown do not take into account the paired nature of the data The number of experiments is shown in parentheses FA represents fatty acids

Tolbutamide effect $CO_2 = +0\ 34 \pm 0\ 16$
Fatty acids $= -0\ 46 \pm 0\ 25$

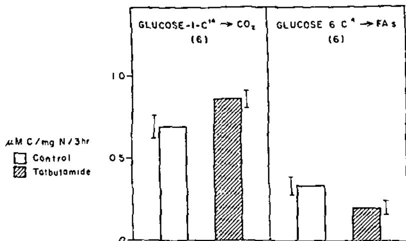


FIGURE 5 Effects of tolbutamide (0 8 mg /ml) upon rat adipose tissue *in vitro* The standard errors shown do not take into account the paired nature of the data The number of experiments is shown in parentheses FA represents fatty acids

Tolbutamide effect $CO_2 = +0\ 17 \pm 0\ 07$
Fatty acids $= -0\ 13 + 0\ 05$

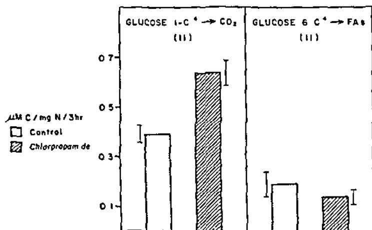


FIGURE 6 Effects of chlorpropamide (0 8 mg /ml) upon rat adipose tissue *in vitro* The standard errors do not take into account the paired nature of the data The number of experiments is shown in parentheses FA represents fatty acids

Chlorpropamide effect $CO_2 = +0\ 21 \pm 0\ 03$
Fatty acids $= -0\ 05 \pm 0\ 03$

acute administration of tolbutamide Recently we investigated the effects of tolbutamide upon the metabolism of glucose by rat adipose tissue and observed (FIGURE 4) that the presence of tolbutamide (0 8 mg /ml) *resulted* in increased CO_2 production from uniformly labeled glucose but in decreased lipogenesis from the same substrate This combination of effects seemed rather unusual since as a rule increased glucose oxidation is also accompanied by increased synthesis of fatty acids from glucose carbon [11, 15, 16] When the observations were repeated with glucose 1 C^{14} and glucose 6 C^{14} as substrates (FIGURE 5) it appeared that the major contributor to the increased CO_2 production from glucose was carbon 1 suggesting stimulation of the phosphogluconate oxidative pathway [17] while lipogenesis again was impaired Finally FIGURE 6 demonstrates a clear cut stimulation of the oxidation of carbon 1 of glucose to CO_2 in the presence of chlorpropamide (0 8 mg /ml) without concurrent stimulation of lipogenesis and indeed with a small inhibition of lipogenesis from glucose 6 C^{14}

Discussion

The observations described simply add two further extrapancreatic effects of the arylsulfonylureas to those already reported on the rat hemidiaphragm [18, 19] on a number of hepatic enzymes related to glycolysis or gluconeogenesis [20, 21] on epinephrine induced glycogenolysis [22] on hepatic transaminase activity [23] and on a number of DPN or TPN-dependent reactions [9] T [illegible] compounds [illegible] described P [illegible] production or increased fatty acid synthesis from acetate associated with decreased ketogenesis in the presence of acetate 1 C^{14} as the substrate Neither the synthesis of cholesterol nor the activity of the reactions leading to the production of acetoacetate from acetoacetyl coenzyme A however has as yet been measured under these conditions It may be of some interest to point out that the observations relating to glucose oxidation and to lipogenesis from glucose by adipose tissue are in accord with the only attempt as yet made to find a common denominator for most or many effects of the arylsulfonylureas on enzyme systems Wallenfels *et al* [9] pointed out that the enzymatic reactions affected by these compounds are frequently dependent upon pyridine nucleotides as cofactors Indeed they have even suggested a hypothetical mechanism of insulin release on this basis The combination of increased CO_2 production from carbon 1 of glucose with decreased fatty acid synthesis is compatible with the presence of some interference in the transfer of hydrogen from glucose 6 phosphate to fatty acids by way of reduced TPN A similar mechanism has been postulated by Cahill and his collaborators[24] for the effects of methylene blue and certain other hydrogen acceptors in liver slices

Summary

Two extrapancreatic metabolic effects of tolbutamide and chlorpropamide have been described First both substances have been shown to inhibit

animals The mechanism by which this antiketogenesis is brought about is unknown Second both substances affect glucose oxidation and lipogenesis from glucose by surviving rat adipose tissue in a manner suggesting interference with the proper transfer of hydrogen from glucose to the fatty acid chain perhaps at the level of the transferring pyridine nucleotides The second observation is in accord with the generalization proposed by Wallenfels for a possible mechanism of action of the arylsulfonylureas upon tissues The relevance of the two effects described to the hypoglycemic action of the arylsulfonylureas in man is unknown

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INHIBITION OF THE DIABETOGENIC ACTION OF SOMATOTROPIN BY TOLBUTAMIDE*

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The mechanism whereby the daily administration of diabetogenic extracts of the anterior pituitary gland or of purified growth hormone (somatotropin) to the dog results in the development of a temporary diabetic state (idiohypophyseal diabetes) that is followed by a permanent diabetic state (metahypophyseal diabetes) is still unknown. A good deal of evidence can be marshalled in support of the hypothesis that somatotropin induces an increased peripheral need for insulin, with a resultant increase in the production of insulin by the beta cells of the pancreas.[1,2] Accordingly, it has been proposed that when the rate of insulin utilization exceeds the rate at which it is manufactured, the beta cells are stimulated excessively and undergo degranulation, hydropic degeneration, and atrophy. Thus the idiohypophyseal diabetes is attributed to a relative insufficiency in the capacity of the pancreas to meet the increased peripheral requirements for insulin, while the metahypophyseal diabetes that persists after the cessation of the somatotropin injections is attributed to the exhaustion, degeneration, and destruction of the beta cells.[1-6]

The same sequence of morphologic changes that is observed in the islet tissue of the somatotropin treated dog[7] is found also during the development of diabetes in the partially depancreatized cat[8] and dog[9] and in the normal or partially depancreatized dog or cat which develops a metadiabetes after the administration of thyroid extracts,[10] anterior pituitary extracts,[4,5,11,12] adrenal corticosteroids,[13] or glucose.[14] Degranulation and various degrees of hydropic degeneration of the beta cells are found also in animals with the idiodiabetes produced during the course of the administration of anterior pituitary extracts or somatotropin,[7,11,12] corticosteroids,[15-17] and large quantities of glucose.[18-20] Concomitant with the degranulation and hydropic degeneration is a reduction in the concentration of extractable insulin in the pancreas.[7,12,21,22]

In accord with the hypothesis that an excessive demand for insulin is a determinant in the production of metadiabetes are the observations that regimes that reduce the peripheral requirements for insulin may also reduce the severity of the idiodiabetes and prevent the development of the metadiabetes produced by various diabetogenic agents.[23] Fasting, undernutrition, a high fat diet, and treatment with insulin appear to reduce the require-

* The work reported in this paper was supported in part by Grant A 1409 from the National Institute of Arthritis and Metabolic Diseases, Public Health Service, Bethesda, Md.

ments for insulin and thereby reduce the production of insulin by the pancreas [24,27] These measures inhibit the effect of diabetogenic factors in varying degrees Thus the changes in the morphology of the islets of Langerhans and the metadiabetes that may occur in partially depancreatized animals are inhibited or corrected by undernutrition[28] and by the administration of insulin [29] Likewise the diabetogenic action of anterior pituitary preparations is diminished or inhibited by undernutrition high fat diets and the administration of insulin [11, 14, 23, 30]

Although it is generally agreed that the degranulation of the beta cells produced by diabetogenic agents is due to excessive stimulation of the production and or release of insulin by the beta cells the significance of the hydropic degeneration in the development of metadiabetes remains unknown The observations of Toreson[31] and of Duff and Toreson[32] established that the morphologic changes identified as hydropic degeneration' reflect the deposition of large amounts of glycogen in the beta cells These studies were extended by Lazarus *et al* [7, 33] who employed a variety of diabetogenic agents and concluded that hydropic degeneration is not due to exhaustion of the beta cells but is a facet of the increased glycogen deposition in cells such as occurs in the kidney and heart of the diabetic animal These studies however do not negate the hypothesis that excessive stimulation of the beta cells is responsible for their exhaustion and atrophy

Irrespective of the precise mechanism whereby metadiabetes is produced by somatotropin it is now quite evident that stimulation of the beta cells and the release of insulin into the circulation is an early effect of its action [1, 3] Thus the administration of purified somatotropin (2 to 3 5 mg /kg /day) to dogs for 6 to 8 days results in an extensive degranulation of the beta cells and a reduction in the concentration of extractable insulin in the pancreas this may occur even before the development of hyperglycemia [7] The extent and the rapidity with which the degranulation occurs is similar to that produced by a number of agents that do not produce a permanent diabetes [33]

lycemic action of isopropylthio the action of this and related sulfonamide derivatives is dependent upon the presence of more than one tenth to one fifth of the pancreas Since the introduction of carbutamide and tolbutamide as orally effective hypoglycemic agents it has been firmly established that their hypoglycemic action is dependent upon the presence of islet tissue capable of producing insulin Although it is quite probable that a number of mechanisms are involved in the action of these agents there is ample evidence that one of these mechanisms is the stimulation of the beta cells and the release of insulin from pancreatic stores [35, 36]

Of particular interest are the observations that the administration of

in the pancreas That

physiological effects that are somewhat similar to those produced during the early stages of the administration of somatotropin is suggested by the dimin

ished glucose tolerance that occurs in rats and dogs treated with carbutamide for a prolonged period [37] In accord also is the demonstration that the usual hypoglycemic phase of the response to the injection of alloxan does not occur in rats given large doses of tolbutamide for 3 or more days [41]

Although some of the early effects of somatotropin appear to be similar to those produced by the sulfonylureas the mechanisms involved appear to be quite different Whereas the excessive stimulation of the beta cells that occurs in the dog treated with somatotropin is probably secondary to an increase in the peripheral requirements for insulin that resulting from the administration of the sulfonylureas is probably due to a direct effect on the islets Thus the insulin requirements of the completely depancreatized dog are increased by somatotropin[1 2 43] and not by the sulfonylureas [44 48] Nevertheless the similarity in the morphologic changes produced by these agents prompted a study on the effect of somatotropin on the acute hypoglycemic action of a single injection of tolbutamide and on the effect of the daily oral administration of tolbutamide on the diabetogenic action of somatotropin

A purified preparation of bovine somatotropin* was given to male dogs weighing from 10 to 17 kg The hormone was injected intramuscularly in a daily dosage that varied from 3 to 10 mg kg The animals were given a constant daily diet and any food not eaten by 4 30 P M was removed and weighed Between 8 and 9 every morning the animals were weighed venous blood samples were taken and somatotropin was injected following these procedures the dogs were fed In some instances blood samples for sugar determination were taken at intervals throughout the day The daily urine excretion was collected quantitatively The concentration of sugar in the blood and urine was determined by Nelson s method [49]

One group of somatotropin treated animals was given a single intravenous injection of tolbutamide† (25 mg /kg) at various intervals before during and after the course of injections of somatotropin Samples of blood were drawn from the fasted animals at intervals before and for 3 hours after the injection of the tolbutamide On the day of the test the administration of somatotropin was withheld until the test was completed A second group of dogs was similarly treated with somatotropin and in addition, was given a single oral dosage of tolbutamide (50 to 100 mg /kg /day)

The administration of somatotropin (3 to 8 mg /kg /day) for from 4 to 6 days results in a change in the character of the hypoglycemic response to the intravenous injection of a standard dose of tolbutamide Whereas the normal dog responds to tolbutamide within 30 min with a precipitous decrease in the blood sugar concentration the animal treated with somatotropin for 4 to 6 days responds with a relatively slow progressive decrease The inhibition of the first phase of the normal acute response to tolbutamide may occur even before the hormone has produced a significant hyperglycemia [50]

* The somatotropin used in this study is a growth hormone preparation provided by the Endocrinology Study Section National Institutes of Health

† We are indebted to The Upjohn Company Kalamazoo Mich for supplies of tolbutamide (Orinase)

the period of treatment The response of the dogs that were given bot somatotropin and tolbutamide, however, differed from the above in that onl a relatively moderate increase in the blood sugar concentration occurred The difference between the responses of the two groups of animals is stati tically highly significant ($p < 0.001$)

Illustrative of the responses of individual animals comprising each grou of dogs are the data depicted in FIGURES 3 and 4 By the fifth day dog 3

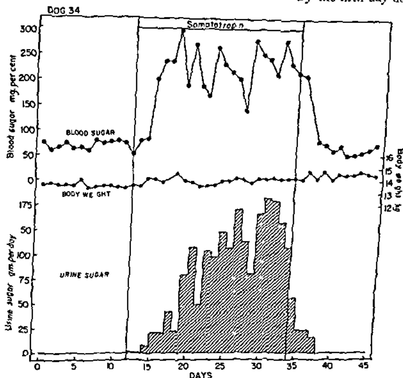


FIGURE 3 The effect of tolbutamide on the diabetogenic action of somatotropin Dog 34 was given intramuscular injection of somatotropin (6 mg /kg /day) throughout the designated period

(FIGURE 3) developed a significant hyperglycemia and glycosuria that persisted throughout the period during which somatotropin was injected Dog 29 (FIGURE 4) however received tolbutamide and developed a hyper

siveness to the diabetogenic action of the somatotropin experi designed in which each animal served as its own control Six dogs were given 2 courses of injections with somatotropin, 3 received tolbutamide orally during the first course, and 3 were given the tolbutamide during the second course of injections *Illustrative of the first group of dogs are the*

data on dog 31 illustrated in FIGURE 5 In this instance the tolbutamide was not started until a definite hyperglycemia and glycosuria were present Although the blood sugar remained above the normal concentration it was significantly lower than that produced in the same dog when it was not given *any tolbutamide during the second course of injections* which was started after an interval of 13 days

That the relatively short interval between the 2 courses of somatotropin injections is not responsible for the more marked diabetogenic response to the second course of injections and for the development of metadiabetes in dog

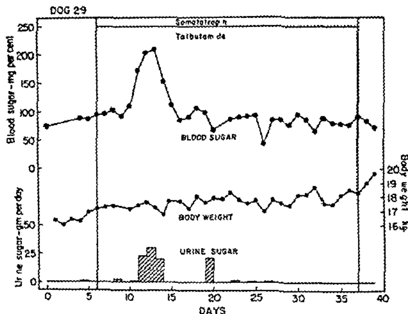


FIGURE 4 The effect of tolbutamide on the diabetogenic action of somatotropin Dog 29 was given the same dosage of somatotropin (6 mg kg day) as dog 34 (see FIGURE 3) plus tolbutamide (50 mg kg day orally)

31 (FIGURE 5) is indicated by the data on dog 38 (FIGURE 6) This dog was given intramuscular injection of somatotropin (8 mg kg day) and after a significant hyperglycemia and glycosuria developed the animal was given tolbutamide (100 mg kg) on the days indicated The blood sugar fluctuated between normal and hyperglycemic levels and a moderate degree of glycosuria occurred The second course was started *340* days after the first course of injections of somatotropin was stopped *It is quite apparent that* in spite of the lengthy interval between the 2 courses of injections a much more marked hyperglycemia and glycosuria occurred during the second course and the animal developed a metadiabetes

In complete agreement with the above are the data obtained in the experi

ments in which the tolbutamide was given during the second course of injections of somatotropin Typical of these are the data illustrated in FIGURE 7 Whereas a significant hyperglycemia and glycosuria developed during the course of injections of somatotropin, a marked inhibition of the diabetogenic response occurred *during the second course*, when tolbutamide was given daily by mouth In no instance did a metahypophyseal diabetes develop after this second course of injections

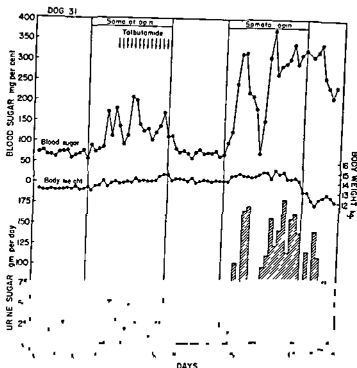


FIGURE 5 Effect of tolbutamide on the diabetogenic action of somatotropin During the first course of intramuscular injections of somatotropin (5 mg /kg /day) tolbutamide (100 mg kg) was given orally at designated days (arrows) A metahypophyseal diabetes followed the cessation of the second course of injections of somatotropin

Illustrative of another experimental design that was used in this study are the data depicted in FIGURE 8 Five days before beginning the administration of somatotropin (5 mg /kg /day), dog 37 was started on an oral dose of tolbutamide (25 mg /kg /day) The tolbutamide was increased to 50 mg kg day on the tenth day after the first injection of somatotropin During the interval a hyperglycemia developed gradually Within 2 days after the tolbutamide dosage was increased, the blood sugar concentration began to fall and reached the pretreatment level by the fifth day On the twenty sixth day after the first injection of somatotropin (that is, 11 days after the blood sugar concentration reached the control level) the tolbutamide

was discontinued but the injection of somatotropin (5 mg/kg/day) was continued for another 27 days. The blood sugar remained unchanged throughout this period. When the dosage of somatotropin was increased to 10 mg/day there was no change in the fasting blood sugar concentration even though there was a higher concentration of glucose in the samples taken at 4 P.M. With the injection of somatotropin (15 mg/kg/day) the fasting blood sugar increased and remained elevated even when the dosage was

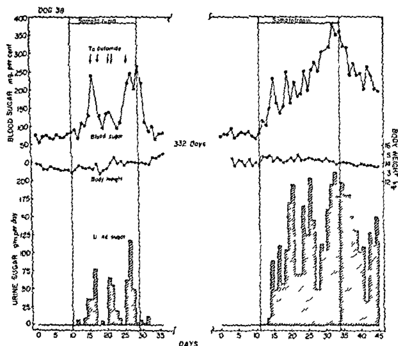


FIGURE 6. Effect of tolbutamide on the diabetogenic action of somatotropin. During the first course of intramuscular injections of somatotropin (8 mg/kg/day) the animal was given tolbutamide (100 mg/kg) orally at designated days (arrows). A metahypophyseal diabetes followed the cessation of the second course of injections with somatotropin.

reduced to 10 mg/kg/day. With the cessation of somatotropin injections the blood sugar returned to normal levels.

The observations reported herein establish the fact that the administration of appropriate quantities of tolbutamide decreases or inhibits the diabetogenic response to the intramuscular injection of somatotropin. Such an effect could not be anticipated on the basis of the morphologic changes in the pancreas that follow the administration of each agent. As noted above, each agent stimulates the pancreas and induces degranulation and hydropic degeneration of the beta cells. If such stimulation and the resultant morphologic changes play a role in the subsequent destruction of the islet

tissue then the concomitant administration of both agents would be expected to enhance the rate of beta cell destr ct on and
of metadiabetes The
validity of the concept
stimulation of the pancreas, with a consequent exhaustion and degeneration of the beta cells

It can be postulated that the tolbutamide induces a discharge of insulin by direct stimulation of the pancreas and that the resultant increase in circulating hormone nullifies the effects of an increase in peripheral requirements

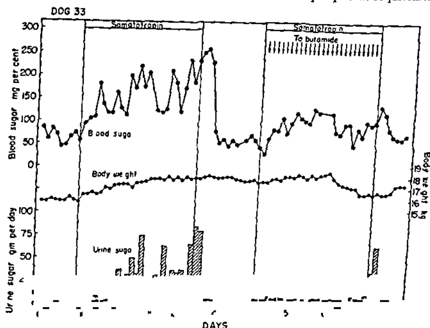


FIGURE 7 Effect of tolbutamide on the diabetogenic action of somatotropin During the second course of intramuscular injections of somatotropin (5 mg /kg /day) tolbutamide (50 mg /kg /day) was given orally

for insulin thereby obviating the diabetogenic action of somatotropin The observations illustrated in FIGURE 1 however suggest that tolbutamide is ineffective in producing a further stimulation of the discharge of insulin after the first few days following the onset of somatotropin injections Further blood samples taken at various intervals during the day from animals treated with both somatotropin and tolbutamide usually showed that the fasting blood sugar was the lowest of the day

An alternative hypothesis is suggested by Anderson and Long s[42] observation that the perfused pancreas stops its secretion of insulin when a growth hormone preparation is added to the perfusate Thus somatotropin may exert some direct action on the beta cells that inhibits their secretory activity Accordingly, the effect of tolbutamide could be attributed to an action in the

beta cell that protects some intracellular mechanism which is vulnerable to the action of somatotropin It is difficult however, to postulate a mechanism whereby such a protective action could be effective for as long a period as that observed in dog *37* However the observation that pretreatment with tolbutamide results in an amelioration of the diabetes that is subsequently produced by the injection of alloxan[43] supports the possibility that tolbutamide exerts some protective action in the beta cells Further clarification may come with similar and more extensive studies with other diabetogenic agents

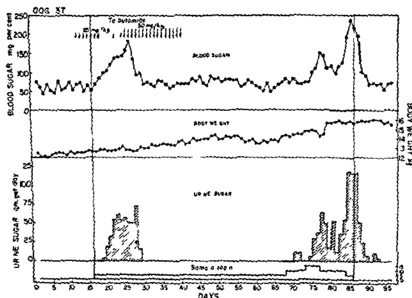


FIGURE 8 Effect of tolbutamide on the diabetogenic action of somatotropin (see text

The data presented herein offer no new insight to the mechanism responsible for the diabetogenic action of somatotropin They do suggest that the growth onset type of diabetes mellitus is not due to the action of somatotropin In contrast with the effect of tolbutamide on the somatotropin treated dog the diabetic child who is responsive to tolbutamide at the onset of the syndrome usually loses his responsiveness with the passage of time irrespective of whether he is treated with tolbutamide[43] or with insulin[44] The patient with acromegaly however who develops diabetes should be responsive to tolbutamide until such time as the beta cells of his pancreas are destroyed Accordingly it is probable that an idiohypophyseal diabetes is responsible for the hyperglycemia of patients with acromegaly who have been found to respond to tolbutamide[45] The prophylactic action of tolbutamide in such patients must await further study

Summary

The intravenous injection of a single dose of tolbutamide (25 mg/kg) to normal dogs results in a precipitous decrease in the blood sugar concentration within 30 min Within a few days after the beginning of the daily intramuscular injection of somatotropin the injection of a similar dose of tolbutamide results in a gradual decrease in the blood sugar Subsequently with the continued administration of somatotropin the hypoglycemic response to the injection of tolbutamide is completely inhibited

The diabetogenic effect of the daily intramuscular injections of somatotropin is decreased or inhibited by the concomitant oral administration of tolbutamide

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DEGRADATION OF INSULIN-I^{131} AND GLUCAGON-I^{131} AND FACTORS INFLUENCING IT*

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As recently reported[1,2] insulin is rapidly degraded in the body and apparently before some of the hormone has exerted its action A better understanding of the factors influencing the degradation of this hormone conceivably could be of assistance in controlling diabetes In this paper are reported results of some of our studies of the capacity of slices and homogenates of most of the body tissues to degrade insulin I^{131} Also are reported the influence on this hormone's degradation of infection trauma, and many factors that presumably influence the amount of insulin secretion, including starvation, phlorhizination, alloxan diabetes prednisolone, insulin hypophysectomy, adrenalectomy or diets high in carbohydrate, fat, or protein

Since glucagon also exerts a powerful influence on carbohydrate distribution and, indeed may cause diabetes,[3] certain phases of the metabolism of this hormone are reported

METHODS

For these studies insulin and glucagon labeled with I^{131} were used Since we have reported many previous observations[1 4 5] with these labeled compounds, details of the techniques employed are discussed in this paper only very briefly However the following points need particular emphasis Great caution must be exercised in the iodination of the hormones because heavy iodination and/or radiation will abolish their chief biological activity and alter their chemical and physical behavior With the iodination techniques employed in our preparations full biological activity can be preserved However, there still tends to be some variation from one batch to the next in some of the chemical properties, necessitating the performance of a sufficient number of control experiments with each batch For the minimization of the adsorption of the hormones to glassware and for completeness in the precipitation of the hormone it is desirable to have at least several milligrams of protein in the container, we commonly used normal plasma In order to work with the most satisfactory ratios of enzyme to substrate we routinely added the corresponding nonlabeled hormone

All insulin I^{131} used was purchased from the Abbott Laboratories, North Chicago, Ill, and was dialyzed at 4° C for several hours immediately before use An insulin-degrading enzyme preparation was used and consisted either of tissue homogenate (liver in most instances) or of the dried powder remaining after special treatment of beef liver homogenate with acetone and ether

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The portion of 4 mg of the extracted liver that was soluble in 0 5 ml of buffer or a specific amount of tissue homogenate was placed in a beaker Either 0 067 M phosphate or, more often, 0 1 M tris (hydroxymethyl)amino methane (Tris), containing 5×10^{-3} M Versene was used as a buffer To the enzyme preparation was added 0 4 μg of insulin I^{131} and 0 1 mg of amorphous insulin, in this system the quantity of enzyme is the limiting factor in the rate of the reaction The total 2 ml mixture, with the *p*H adjusted to 7 4, was placed in a Dubnoff metabolic shaker at 37° C for 5 min *Fifteen seconds before the end of the incubation 1 ml of 2* per cent of plasma in water was added to ensure complete precipitation of insulin, at the end of *the incubation period 3 ml of 20 per cent trichloracetic acid (TCA)* was added After standing for 30 min the mixture was centrifuged The solid residue was washed 2 to 5 times with 10 per cent trichloracetic acid, and the washings were added to the supernatant Each sample was run in triplicate

The radioactivity in each specimen was measured by a well type gamma counter The precipitate was dissolved in 30 per cent KOH and brought to a volume equal to that of the combined supernatants A calculation was then made of the percentage of the total *radioactivity found in the supernatant* fractions this fraction was regarded as an indicator of the amount of degraded hormone At least the following 2 control studies were conducted simultaneously with each experiment (1) 1 ml of buffer plus 1 ml of insulin solution, and (2) 0 5 ml of buffer plus 0 5 ml of enzyme preparation plus 1 ml of insulin solution The exceptions to the foregoing technique are described where indicated

All rats were male and, except as noted later, were of the Sprague Dawley strain

Insulin I^{131} Degradation by Homogenates of Different Tissue

Nonfasted rats
approximately 3(
gastrocnemius
*p*H 7 5 and homogenized *From 1 to 15 per cent concentrations of the homogenates* were incubated with insulin I^{131} (0 5 μc , 1 μg) and 13 μg of amorphous insulin The amount of degradation was determined as described under Methods

As seen in table 1, there was an increase in the degradation of insulin I^{131}, with an increase in the concentration of homogenate used In concentrations from 1 to 7 5 per cent the effect was essentially linear, in spite of the fact that the experiments were not conducted simultaneously or with the same batch of insulin I^{131} Liver and kidney were much more active *than* gastrocnemius muscle

Our studies were extended to determine to what extent other tissue homog

0 15 ml was used in each vessel

The results are shown in table 2 Since 6 different experiments were con-

TABLE 1
INSULIN I131 DEGRADATION BY DIFFERENT CONCENTRATIONS OF TISSUE HOMOGENATES

Experiment No *	No tissue specimens	Concentration of homogenate (per cent)	Average % insulin I131 degraded		
			Liver	Kidney	Muscle
1	4	1	13 1	9 5	6 6
	4	3	32 8	20 5	5 0
2	5	5	32 1	30 1	11 5
3	5	7 5	62 3	51 3	19 7
	5	15	74 4	64 3	25

* The 3 experiments were conducted at intervals separated by weeks and different batches of insulin I131 were used

ducted, with different batches of insulin I131 and with animals of various sizes at widely separated intervals all of the results are recorded with reference to the diaphragm, to which is assigned a value of 100 It is seen that essentially all of the tissues caused some degradation of the hormone Among the more active tissues were liver, pancreas, kidney, and testis On the other hand plasma had no activity, and washed red blood cells had essentially none The action of brain, diaphragm gastrocnemius and lymph node was relatively small in the case of muscle however, its body mass is so great that its total action is very significant

INSULIN I131 DEGRADATION BY SLICES OF DIFFERENT TISSUES

[illegible]

Approximately 200 mg of each of these tissues was obtained from each of 12 male rats For each 100 mg of tissue 1 25 μg of insulin I131 was added, as well as 10 μg of amorphous insulin Upon completion of the incubation which was conducted as with the homogenates, the medium was poured into 1 ml of 2 per cent plasma Ten ml of 20 per cent TCA was then added, the precipitate was washed twice with 2 ml of 5 per cent TCA The tissue was washed 4 times with 2 ml of saline The washings were combined Counts of radioactivity in the supernates and washes were as described earlier

EFFECT OF INFECTION AND TISSUE DAMAGE ON INSULIN DEGRADATION

In view of the fact that, first, the daily insulin requirements for diabetics are greatly increased with infection and trauma, and second, essentially all

TABLE 2
INSULIN I[131] DEGRADATION BY VARIOUS RAT TISSUE HOMOGENATES*

Experiment No †	1			2		3		4		5		6	
Rat No ‡	1	2	3	4	5	6	7	8	9	10	11	12	13
Liver						464	423	570	657	616	761	249	252
Pancreas	251	257	264	278	299	376	328	364	414	859		736	582
Kidney										516	540	217	231
Kidney cortex	217	235	265	252	261	410	335	362	419	458	619	178	227
Testis	291	264	282	291	295	458	412	376	408	325	419	114	149
Small intestines	178	299	258	225	216	131	131	191	169	248	322	135	165
Stomach	141	188	144	191	199	329	232	204	237	460	825	384	337
Spleen	189	169	190	199	205	361	342	248	314	348	543	172	139
Heart	*139*	*174*	*141*	*109*	*135*	*343*	*309*	*273*	*237*	159	194	70	96
Diaphragm	100	100	100	100	100	100	100	100	100	100	100	100	100
Gastrocnemius muscle						*143*		*84*	*119*	*75*	*87*	*45*	*45*
Brain	102	94	101	110	100	222	249	115	108	45		19	12
Adipose	54	52	54	*27*	*33*	*113*	96	88		*22*	30	*22*	*21*
Thymus	149	145	132	164	178								
Lymph nodes	102	81	71	80	92								
Red blood cells	5	2	1	4	7								
Plasma	0	0	0	0	0								

* All values are related to the action of diaphragm which was arbitrarily given a value of 100

† The various experiments were conducted over an interval of 3 years (see text)

‡ In rats 1 to 9 100 to 150 mg of tissue was used per beaker in rats 10 to 13 15 mg in all instances the ratio of tissue weight to insulin weight was 10 1 In the experiments where only 15 mg of tissue homogenate was used 100 mg of bovine plasma albumin was added in order to reduce adsorption of the hormone to glass

tissues can degrade insulin we were prompted to ascertain whether increased insulin degradation was demonstrable under these circumstances

Effect on Degradation of Insulin I^{131} in Vitro of Serum from Diabetics with Infection

As previously mentioned normal serum has been shown not to degrade insulin, but it was considered possible for the serum of infected diabetics to act differently Therefore through the courtesy of Joseph Crampton serum was obtained from 2 diabetics each of whom was suffering from a carbuncle the development of which caused the need for about twice the usual daily dosage of insulin Incubation of 1 ml of serum with insulin I^{131} for 1 hour like that of serum of 4 normals studied simultaneously, caused no degradation of the hormone

Degradation of Insulin I^{131} in Diabetics and Nondiabetics with Infection

Insulin I^{131} was injected intravenously as described by Welsh *et al* [7] and plasma samples were obtained 1 hour later from 11 diabetics and 8 nondia

betics with infection Approximately one half of each group of subjects had fever at the time of the test, in a few patients the infection was severe (for example, pneumonia) The amount of TCA precipitable radioactivity in the serum 1 hour after insulin I131 administration was compared with large series of noninfected diabetics and nondiabetics [7] There was no evidence that the infections were associated with increased degradation In 2 diabetic patients the test was repeated when the subject was free of infection In one, the results of the 2 tests were identical, in the other, there was actually less degradation with the infection because there was more binding to plasma globulin

In view of the fact that there were many variables that might have influenced the insulin degradation, the problem was studied more specifically in animals and *in vitro*

Effect of Staphylococci on Insulin I131 Degradation in Vitro

Because of the fact that staphylococci are such common offenders in diabetics, a total of 5 *in vitro* experiments was conducted with 2 highly virulent strains of this organism

Experiment I A heavy broth culture of staphylococci was centrifuged, washed twice with saline and then suspended in saline One half of the material was subjected to supersonic vibration for 45 min to effect cellular disruption The material was then recentrifuged, and part of the supernatant was tested as such whereas another part was boiled before testing Thus there were 3 preparations of washed staphylococci (1) whole cells (2) supernatant from disrupted cells and (3) a portion of the second preparation that had been boiled Each preparation was incubated for 5 min with insulin I131 and 4 mg of amorphous insulin alone or with the insulins plus liver insulinase As the results shown in TABLE 3 indicate none of the preparations of staphylococci caused insulin degradation Indeed the boiled staphylococcal supernatant actually decreased the degradation by the insulinase This may have resulted from increased binding of insulin I131 to the boiled staphylococcal products

TABLE 3
EFFECT OF STAPHYLOCOCCI ON INSULIN I131 DEGRADATION

Insulin I131	Liver enzyme	Other	Per cent degradation*
+			1 1
+	+		12 7
+		Washed staph	1 1
+	+	Washed staph	13 0
+		Sonified staph	1 4
+	+	Sonified staph	11 6
+		Boiled sonified staph	1 4
+	+	Boiled sonified staph	7 3

* Average of triplicate determinations

Experiment 2 In this experiment the incubation was conducted at pH 6 0 and 7 5, only the latter was tested in the previous experiment In this experiment there was used 80 mg (dry weight), per incubation flask, of staphylococcal products in the supernatant and 16 mg of the whole organisms Again, the staphylococci caused no degradation

Experiment 3 The staphylococci in this and the next 2 experiments were obtained from one of the aforementioned diabetics with carbunculosis and increased insulin requirements The bacterial material was prepared as previously described However the incubations were with only 0 025 as much amorphous insulin and were for 1 hour instead of for 5 min The determinations were at pH 7 5 and 6 0 Again, no evidence of degradation by the staphylococci was observed

Experiment 4 The previous experiment was repeated, using a much greater concentration of staphylococci or their products and with one half as much amorphous insulin No degradation from the bacteria was observed

Experiment 5 Experiment 4 was repeated except that 10 mg of bovine plasma albumin was added to each flask in order to minimize adsorption of insulin I^{131} to glassware and or products in the medium There was no significant change in the results

Effect of Staphylococcal Infection in Rabbits on Insulin I^{131} Degradation by Muscle

All rabbits used were large white males

Experiment 1 Tourniquets consisting of a rubber band and a copper wire were placed high on the right thigh of each of 2 rabbits Seven hours later the tourniquets were removed, and the gastrocnemius as well as surrounding structures were injected with 0 2 cc of a heavy suspension of staphylococci obtained from a diabetic patient, as described earlier Five hours thereafter the animals were killed by a blow on the head About 12 gm of traumatized and staphylococci injected muscle was removed, as well as a comparable amount of muscle from the other hind leg After weighing, this material was homogenized in approximately 40 ml of 0 1 M Tris in a Waring Blendor Following centrifugation the supernatant was removed, and 2 ml was incubated for 30 min with 1 ml of insulin I^{131} containing 5 μg of amorphous insulin The amount of insulin I^{131} degraded was determined in the manner described previously There was no difference in the degradation caused by the muscle extract from the 2 legs

Experiment 2 An experiment similar to that above was conducted with 5 rabbits, but 1 ml of a heavier suspension of the staphylococci was injected into each hind leg The supernatants from the muscle homogenate each contained 22 mg of solids per 1 ml Each incubation flask contained 13 μg of insulin There was no difference in the degradation caused by the various extracts

Effect of Staphylococcal Infection in Rabbits on Insulin I^{131} Degradation by Serum

In the same white male rabbits described in the previous section, serum specimens were studied About 15 ml of blood was withdrawn before the

tourniquets were applied and a few minutes after removing the tourniquets and massaging the leg in the second experiment an additional blood specimen was withdrawn 20 hours after initiation of the experiment Two ml of serum was incubated for 15 min with 4 or 13 μg of insulin in 1 ml Tris along with a small amount of insulin I^{131} None of the serum specimens caused degradation of the hormone

Effect of Staphylococcal Infection in Rats on Insulin I^{131} Degradation in Vivo

Under ether anesthesia each of 6 male rats weighing approximately 250 gm was injected intraperitoneally with 0 5 ml of a heavy suspension of staphylococci obtained from the previously discussed diabetic patient Six days later following a 24 hour fast each rat received 30 μc of insulin I^{131} in a tail vein Thirty minutes thereafter under ether blood was removed from the aorta in a heparinized syringe The amount of degraded and undegraded insulin was measured An average of 49 per cent of the insulin I^{131} was degraded and in another group of 6 rats treated in the same manner except for the injection of staphylococci there was a 50 per cent degradation of insulin I^{131}

Effect in Rats of Infection with Fecal Contents on Insulin I^{131} Degradation in Vivo

Five ether anesthetized rats were injected subcutaneously with 2 cc of a heavy fecal suspension in water Eighteen hours later insulin I^{131} was injected into the tail vein as in the previous experiment The average degradation was 43 per cent whereas in 5 controls treated in the same manner except for the injection of fecal suspension the average degradation was 42 per cent

Effect of Necrosis of a Rabbit Leg on the Capacity of the Serum or Damaged Muscle to Degrade Insulin I^{131} in Vitro

Experiment 1 A rubber band and a copper wire were wrapped tightly around the upper part of the right hind leg of each of 2 rabbits Five hours later the tourniquet was removed and the leg massaged for 2 min Approximately 15 ml of blood was withdrawn from the heart before the tourniquets were applied as well as 2 min and 20 min after their removal To 1 ml of serum was added 4 mg of amorphous insulin 10 μg insulin I^{13} and 2 ml of Tris buffer The mixture was incubated for 1 hour and the amount of insulin I^{131} degraded was measured None of the serum specimens promoted degradation

Experiment 2 Under ether anesthesia the right thighs of 7 rabbits were severely crushed with a hammer This procedure subjected the animals to severe stress as evidenced by the fact that many died The serum from blood drawn before the crushing 2 5 hours and 8 hours later when incubated for 15 min with insulin I^{131} caused no degradation of the latter About 10 gm of muscle was removed from each thigh and homogenized in saline One aliquot of the supernatant from a 25 per cent muscle homogenate was adjusted to *p*H 7 5 with Tris buffer and another to *p*H 6 0 with phos

phate One ml of each of these mixtures was incubated for 15 min with 30 µg insulin and insulin I^{131} in 1 ml of water All tests were conducted in duplicate The amount of insulin I^{131} degraded by the traumatized and nontraumatized muscle extracts was comparable Experiments with the control muscle were conducted using twice or 0 5 the concentration of the muscle extract used above The degradation of insulin I^{131} was proportionate to the amount of extract

Effect of the Crushing of a Rat Leg upon the Degradation of Insulin I^{131} in Vivo

Under ether anesthesia the right hind legs of 6 male rats were severely crushed with a hammer The process was repeated 4 days later and the animals were then fasted for 24 hours Insulin I^{131} was injected into a tail vein and after 30 min blood was removed from the aorta and analyzed for degraded and undegraded hormone as previously described Six other rats were handled exactly in the same manner excluding the trauma The average amount of degraded insulin I^{131} in the controls was 50 per cent and in the traumatized group 55 per cent ($p < 0.001$) Three months later the experiment was repeated with 6 rats in similar manner except that only one traumatization was inflicted 24 hours before sacrifice and the rats were not fasted The amount of degradation was 47 per cent in the traumatized group and 41 per cent in the controls ($p < 0.01$)

Effect on Insulin I^{131} Degradation of Conditions Presumably Influencing Insulin Secretion

Many chemical agents in the body in some manner exert a contrainsulin action Since insulinase markedly degrades insulin we were prompted to investigate in rats the effect on the insulinase action of many individual factors listed in TABLE 4 that influence carbohydrate metabolism and presumably the secretion of insulin In each individual experiment there were 6 rats except for one alloxanized group and the adrenalectomized group each of which included 4 rats Thus there were 116 experimental rats with completed studies and 60 controls The high fat diet consisted of a ratio of 1 gm of peanut oil to 1 gm of ground Purina Fox Chow * The high carbohydrate diet consisted of 3 parts of fox chow to 7 parts sucrose and the high protein diet contained 7 parts casein per 3 parts of fox chow thus each of the 3 diets contained more than 75 per cent of its calories as fat carbohydrate or protein Twelve gm rat/day was served and all was devoured The phlorhizinized and alloxanized rats showed persistent glycosuria Ultralente insulin was injected subcutaneously once daily For the experiments with inactive insulin an ultralente preparation was treated with cysteine Prednisolone acetate was injected subcutaneously once daily As shown in TABLE 4 some rats were of the Sprague Dawley strain and some were Wistars some were fasted and others not some were treated on the day of sacrifice and others not All rats were weighed at the beginning of the experiment (approximately 250 gm) and on the day of sacrifice All of the procedures were rigorous especially the prednisolone and insulin

* Product of Purina Mills Buffalo N Y

TABLE 4

EFFECT ON INSULIN I[131] DEGRADATION BY LIVER HOMOGENATES OF CONDITIONS PRESUMABLY INFLUENCING INSULIN SECRETION

Treatment				Rats			Insulin I[131] degraded									
Type	Days	Daily dose/rat	On day of expt	Fasted	Sprague Dawley	Wistar	μg wet weight		μg dry weight		μg protein		Mg/whole liver		Mg/100 gm body weight	
							Expt*	Con	Expt	Con	Expt	Con	Expt	Con	Expt	Con
Starvation	4				+		5.5	5.3	17.6	16.4	1.5	1.6	(11.8	4.0)	(0.9	1.3)
High fat diet	6		0	+		+	5.9	5.3								
High fat diet	6		+	0	+		6.5	5.3	(20.1	16.4)†	2.1	1.6	3.7	4.0	1.4	1.5
High protein diet	22		0	+		+	5.0	5.3								
High protein diet	10		+	0	+		5.3	5.3	1[illegible]	16.4	1.5	1.6	4.0	4.0	1.4	1.3
High CHO diet	22		0	+		+	6.5	5.3								
High CHO diet	10		+	0	+		5.4	5.3	16.9	16.4	1.9	1.6	3.7	4.0	1.3	1.3
Phlorhizin	13	0.1 gm/kg q.d.	0	+		+	[illegible]	3.8								
Alloxan	35			0		+	4.1	4.4	10.0	10.[illegible]	1.3	1.4	1.9	3.5	(1.6	0.9)
Alloxan	13			0	+		5.6	4.2	(17.6	13.4)	1.6	1.3	3.1	2.9	1.5	1.1
Adrenalectomy	13			0			4.4	4.4	10.0	10.7	1.5	1.4	3.2	3.5	(1.5	0.9)
Hypophysectomy	13			0		+	4.8	4.4	10.0	10.7	1.3	1.4	3.0	3.5	0.9	0.9
Ultralente insulin	7	6 U	0	0		+	2.3	2.3	7.7	6.6	0.7	0.7	1.6	1.9	0.6	0.5
Ultralente insulin	6	6 U	+	0	+		(5.1	3.8)	16.8	12.2	1.3	1.2	3.1	2.7	1.1	1.0
Ultralente insulin	6	6 U	+	0		+	2.3	1.8					1.6	1.3	0.5	0.4
Ultralente insulin (inactive)	13	8 U	0	0		+	4.6	3.9					3.2	2.8	1.1	0.9
Ultralente insulin (inactive)	6	60 U	+		+		4.9	3.8	16.1	12.2	1.5	1.2	3.3	2.7	1.2	1.0
Prednisolone acetate	8	5 mg	0	+		+	(3.0	4.5)								
Prednisolone acetate	10	5 mg	+	0	+		3.7	3.8	11.5	12.2	1.1	1.2	2.3	2.[illegible]	1.2	1.0
Prednisolone acetate	8	5 mg	+	0		+	[illegible].7	[illegible]					2.5	2.4	(1.5	0.9)
Prednisolone acetate	8	5 mg	+	0		+	1.4	1.8					1.3	1.3	0.5	0.4

* Expt = experimental animals Con = controls

† The bracketed figures represent statistically significant differences ($p < 0.01$)

administrations A number of rats in these groups died The alloxanized starved and prednisolone treated rats lost a great deal of weight Those on high protein or high fat diets and those phlorhizinized lost a small amount the adrenalectomized hypophysectomized and high carbohydrate group did not gain Each rat was decapitated and the liver rapidly removed washed in 0 1 M Tris buffer blotted on moist filter paper and weighed The liver was homogenized in Tris A 1 ml aliquot containing 15 mg liver (wet weight) was incubated at 37° C with 90 µg of insulin as well as insulin I^{131} in 1 ml Tris for 5 min Fifteen seconds before the end of incubation 1 ml of 2 per cent plasma was added The reaction was stopped by the addition of 3 ml of 20 per cent TCA The mixture was stirred allowed to stand for at least 30 min then centrifuged decanted and the residues washed twice with 5 per cent TCA The washes were combined and the radio activity of the supernatants as well as the precipitates measured as described earlier Since different intervals elapse during the preparation of the liver homogenates studies were first conducted at 4° C to determine such effects on insulinase activity Fifteen rats each weighing about 300 gm were decapitated and at 0 2 4 6 and 8 hours thereafter the livers from 3 animals were removed and the insulinase determined no significant difference was found

Compared with the controls which were of comparable body weight at the beginning of the experiments the total liver weights were significantly less in the following groups adrenalectomy hypophysectomy alloxan high fat and starved they were increased in the prednisolone and high protein groups The percentage of water was decreased in the adrenalectomy hypophysectomy and the prednisolone groups.

The averages of the other major observations are shown in TABLE 4 Considering the marked changes in metabolism there were surprisingly few alterations in liver insulinase activity There was an increase in the degradation of the liver homogenate of one high fat and one alloxan group when the calculations were based on a liver dry weight ratio but not by the other standards On a liver wet weight basis there was one insulin group that had an increased degradation and one prednisolone group that had a [illegible]

weight Considering the radical metabolic changes that were found to be induced by the various procedures the relatively few abnormalities in liver insulinase were surprising

Glucagon I^{131} Degradation

Using glucagon labeled with I^{131} in the manner described previously [8,9] it was found that homogenates of pancreas liver kidney cortex and muscle broke down this labeled hormone Indeed in concentrations of homogenates from 0 05 to 0 4 per cent and 5 µg of glucagon I^{131} the relations were essentially linear (FIGURE 1) Studies with muscle and liver however showed

that the effect tended to plateau when more than 1 per cent concentration was used

Glucagon I^{131} degradation by acetone precipitates from liver homogenate is inhibited by many of the same compounds that inhibit the insulin I^{131} degradation[10, 11] However there was a different degree of inhibition in some instances For example the following inhibited insulin I^{131} degradation more than glucagon I^{131} benzoyl L tyrosine ethyl ester acetyl L tryptophan ethyl ester tannic acid dithiouracil bis (dimethylcarbamyl)

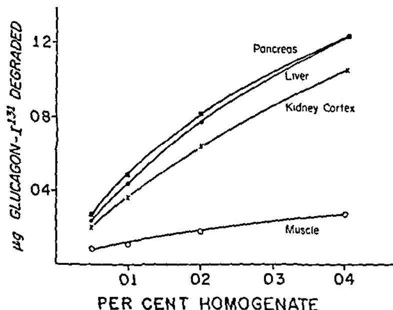


FIGURE 1 The degradation of glucagon I^{131} by varying concentrations of tissue homogenates

disulfide and 1 butyl 3 *o* aminophenylsulfonylurea A comparable degree of inhibition of degradation of the 2 hormones was observed with phenethyl biguanide and 1 phenyl 3 phenylsulfonylurea

Discussion

Before discussing our results consideration will be given to some of the biochemical factors involved in insulin and glucagon degradation It has long been known that glutathione (GSH) and other such reducing substances inactivate insulin[12] indeed the hormone is inactive when only one third of the disulfide linkages have been reduced[13, 14] Insulin is inactivated

could be glutathione since it definitely degrades insulin I^{131}, however,

reduction is not a prerequisite for insulin I^{131} degradation because an acetone precipitate of liver homogenate degrades insulin I^{131} even after dialysis for 48 hours[17] Such a preparation causes much less degradation than an undialyzed one or a dialyzed one to which glutathione has been added,[18] under certain conditions glutathione activity synergizes enzymatic degradation and conversely, enzymatic action stimulates degradation by glutathione[18] Rat liver has been shown to contain an enzyme designated as glutathione reductase[19] that catalyzes the reduction of oxidized glutathione (GSSG) This TPN-linked GSSG reductase system does not act directly on insulin but promotes regeneration of GSH that has been oxidized by insulin it increases the effectiveness of a given amount of glutathione There is evidence suggesting that there is a second enzyme in the liver that directly reduces insulin probably by promoting transfer of hydrogen from GSH to

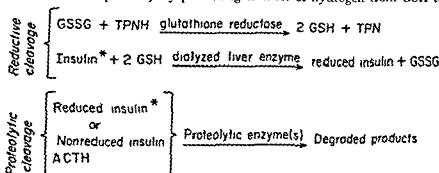


FIGURE 2 Liver enzymes that may be involved in the degradation of insulin and other hormones Somatotropin prolactin vasopressin or oxytocin also may engage in the reactions indicated by the asterisks

insulin[18] These observations are in keeping with a preliminary report by Racker[20] suggesting that enzymatic reduction of insulin by liver might occur In conclusion as diagrammed in FIGURE 2 it appears that reduction of an enzymatic and nonenzymatic type causes the inactivation and degradation of insulin However this presumably is not the only method of degradation, because many observations have suggested[1,2] that the liver contains one or more enzyme systems that cause proteolytic degradation of insulin The use of insulin I^{131} has been of great assistance in these studies[2,8,21,22] Mirsky has presented the hypothesis that there is a single enzyme in liver that has a high degree of specificity for the degradation of insulin[22] This may prove to be the case, but it has not been possible to establish it as a fact because of the crudity of the enzyme preparations that have always been used[24] such preparations probably contain several enzymes and act on a number of substrates other than insulin Tomizawa and Halsey[25] have recently isolated a protein fraction from beef liver apparently pure by starch block electrophoretic and ultracentrifugal patterns, which degrades insulin I^{131} However glucagon I^{131} degradation is not demonstrated by the usual method The specific mechanism of degradation of insulin is under investigation

In the studies conducted with crude liver extract it has been shown that the degradation of insulin I^{131} is decreased by casein α corticotropin glucagon somatotropin and certain other proteins [24] On the other hand other proteins for instance albumin [22, 24] exert no effect Actually many protein hydrolyzates and amino acids as well as a vast number of other compounds inhibit insulin I^{131} degradation [2, 10, 1, 26, 27] some of the inhibitors are competitive and some are noncompetitive The same crude liver enzyme preparation that degrades insulin I^{131} has been shown to degrade glucagon I^{131} also [8, 9, 29] The degradation of each hormone is also inhibited by α-corticotropin α casein and somatotropin nonlabeled glucagon or insulin [8, 29] phenethylbiguanide 1 butyl 3 o aminophenylsulfonylurea 1 phenyl 3 phenylsulfonylurea bis (dimethylcarbamyl) disulfide dithiouracil tannic acid acetyl L tryptophan ethyl ester benzoyl L tyrosine ethyl ester p chloromercuribenzoic acid and many other compounds including hydrolyzates of the aforementioned proteins [28] The fact that the enzyme action is blocked by p-chloromercuribenzoic acid and other agents that react with sulfhydryl
action T
insulin I^{13}
enate subjected to differential centrifugation [8, 29] Each labeled hormone is broken down by most ti sues of the body [1, 9] the liver and kidney being particularly active

In spite of the many similarities of enzymatic degradative actions on insulin and glucagon a number of differences have been demonstrated For example GSH greatly increases the amount of degradation of insulin I^{131} caused by a dialyzed liver enzyme preparation whereas it has little effect on glucagon I^{131} degradation Iodoacetate and cupric ion agents that react with glutathione inhibit insulin degradation more than glucagon [30, 31] Aqueous or bicarbonate extracts of rat liver lose their insulin hydrolyzing action on standing overnight in the cold on dilution or on blending for 2 min these procedures do not have much effect on the amount of glucagon degradation [9] Citrate increases insulin degradation but does not affect that of glucagon [29, 32] One liver preparation has been reported[33] to inactivate glucagon but not insul n Conversely as mentioned earlier the purified enzyme isolated by Tomizawa and Halsey degrades insulin I^{131} but not glucagon I^{131}

In analyzing the studies reported thus far it appears (FIGURE 2) that in insulin degradation enzymatic and nonenzymatic reductions are important whereas the e reduction proces es are not involved in glucagon degradation since glucagon does not contain disulfide bonds However proteolysis seems to occur in the degradation of each hormone [19, 2, 34] We do not know how and *to what extent the reductive and the proteolytic reactions are* related It is possible that other d sulfide-containing hormones such as somatotrop n prolactic oxytocin and va opre in may like in ulin be degraded by both reductive and proteolytic reactions whereas the degrada t on of corticotropin like that of glucagon may involve proteolysis only

As previously noted most tissues have the capacity to degrade glucagon I^{131}

as well as insulin I^{131} in each instance the liver kidney pancreas and testis are among the most active Not much is known about the degradative systems of tissues other than the liver Muscle contains a nondialyzable heat labile system[35] Corticotropin somatotropin oxidized insulin α casein and glucagon inhibit the degradation of insulin I^{131} by muscle and enhance the glucose uptake by rat diaphragm[35] the enhancing effect of glucagon on insulin increased glucose uptake had been noted earlier by Randle[36] Crude extract of beef anterior pituitary also contains an enzyme system that causes insulin I^{131} degradation[37] It has been shown that insulin I^{131} degradation *in vivo* is reduced in hypophysectomized rats[38]

In studies with insulin I^{131} it was shown that tissue slices were much less active than homogenates although the relative order of effectiveness of the different tissues was similar this apparently is due to the liberation of the large quantities of enzyme(s) from the soluble intracellular fraction

Once insulin is bound to protein it is less subject to degradation[7, 39] Indeed the binding of insulin I^{131} to plasma globulin accounts for the abnormally slow degradation of insulin I^{131} in subjects who have had insulin treatment for several weeks or longer[7, 40] diabetics not treated with insulin apparently have a normal rate of insulin degradation Even those with infection were found to have a normal rate of degradation of insulin I^{131} *in vivo* and sera from such patients incubated with insulin I^{131} did not degrade insulin I^{131} Many studies were conducted to investigate further whether the increase in dosage of insulin required by diabetics with infection or trauma was due to increased insulin degradation However there was no degradation produced by sera of patients or animals infected with staphylococci and no increased degradation *in vivo* with staphylococcal infection or in rats infection caused by the subcutaneous injection of fecal suspensions Whole or fragmented staphylococci incubated with insulin I^{131} did not degrade it Severe traumatization of the hind leg of rabbits did not cause the presence of insulin I^{131} degrading material in the serum whether or not there also was infection of the muscle With muscle traumatization in rats there was demonstrated an increase in insulin I^{131} degradation *in vivo*

Because of the fact that insulin I^{131} is degraded very rapidly in the body and because the liver has a large capacity for this action and has the first chances to react with endogenous insulin the quantity of insulinase activity in liver was studied in rats subjected to one of several vigorous procedures thought to influence significantly the amount of insulin secreted These included starvation phlorhizination alloxan diabetes injections of prednisolone or insulin hypophysectomy adrenalectomy and diets high in carbohydrate fat or protein Starvation was previously reported to decrease liver insulinase activity[41] we found that it decreased the total liver insulin I^{131}-degrading capacity A few abnormalities were produced by other factors but we were impressed by the fact that they were relatively small considering the strenuous nature of the procedures These observations raise the question as to how extensively the insulinase system is involved in the homeostasis for carbohydrate metabolism However it always has a potent insulin degrading capacity and there is evidence that

the greater the insulin content in the body [1,2] the larger is the total amount degraded

Summary and Conclusions

Essentially all tissues possess the capacity to degrade insulin I[131] and glucagon I[131] rapidly liver pancreas kidney and testis are among the most active Much more information must be obtained before the mechanism(s) of degradation are established but in the case of liver present information suggests that insulin is degraded by enzymatic and nonenzymatic reduction as well as by proteolysis whereas glucagon degradation is affected by the latter process alone Tissue homogenates degrade insulin much faster than tissue slices Many different compounds inhibit the degradation of insulin and glucagon some by competitive and others by noncompetitive reactions Staphylococcal infections in patients rats and rabbits do not cause the serum to degrade insulin I[131] Indeed neither whole staphylococcal cells nor disrupted cells cause degradation of the hormone Traumatization of the rabbit leg does not cause the serum to degrade insulin nor does it increase the degradative capacity of muscle However traumatization of rat leg causes an increased degradation of insulin I[13] *in vivo* Of many procedures assumed to influence significantly the amount of insulin secreted few change the degradative capacity of liver to a biologically significant degree The conditions tested were starvation insulin or prednisolone injections hypophysectomy adrenalectomy alloxan diabetes phlorhizination and diets high in carbohydrate fat or protein Additional study is needed to determine the role of insulin degrading enzymes in homeostasis and in the pathogenesis of diabetes

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DETERMINATION OF THE RATE OF INSULIN DESTRUCTION *IN VIVO**

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Insulin given by intravenous injection disappears rapidly from the plasma. The half time of insulin labeled with iodine 131 (I^{131}) in the intravascular space has been shown to be approximately twenty five minutes.[1] The rapid decline in plasma concentration is the result of several events that occur simultaneously: first, insulin undergoes proteolytic destruction in the liver and kidney;[2] second, insulin may be bound or sequestered by tissue cells;[3] and third, insulin may be dispersed in the compartments of body water. The experiments reported in this paper were designed to determine the extent to which each of these processes affects insulin disappearance.

Methods

Normal male rabbits were used in this study. Animals were sacrificed at the end of each experiment by intravenous injection of lethal quantities of pentobarbital. Complete urine collections were obtained by excision of the bladder. The experiment was discontinued if spontaneous voiding occurred. Eviscerated animals were prepared by removal of the intestinal tract from the lower esophagus to the rectum, including the spleen and pancreas, under barbital narcosis and ether anesthesia. The arterial blood supply to the liver was ligated and sectioned. Barbital narcosis was continued in these animals throughout the experiment. Nephrectomy was performed either alone or in association with evisceration, as described in the experimental detail.

Isotonic sterile saline was added to radioinsulin† to form a concentration of approximately 5 μc/ml. The solution was stored at 10° C.

At the start of each experiment each animal was given intravenous injection of 2 ml. of a 10 per cent solution of NaI. A 20 min. period of circulation of the NaI was allowed in order to block uptake of free I^{131} by the thyroid gland. Crystalline zinc insulin was then given by intravenous injection to some of the animals. Radioactive insulin was administered intravenously 10 min. after the injection of crystalline zinc insulin. A 50 per cent solution of glucose (1 to 2 ml. I.V.) was used to prevent hypoglycemia.

The volume of injected radioinsulin was measured accurately in a syringe calibrated by means of a threaded brass rod; the plunger of the syringe was advanced by a screw. In order to determine the quantities of insulin injected, an aliquot of the material that had been drawn into the syringe was

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† Insulin I^{131} was obtained from Abbott Laboratories, Oak Ridge, Tenn.

added to 50 ml of pooled human plasma (serum phantom) Analysis of the radioactivity in this serum phantom gave an accurate measure of the characteristics and quantity of radioactive material injected into the rabbit

The analysis of both the serum phantom and of the rabbit serum were carried out by the same technique on 0 1 ml of serum Intact insulin was isolated by electrophoresis or hydrodynamic flow [1,4] The precipitable radioactivity was obtained by adding 1 ml of 10 per cent trichloroacetic acid (TCA) to 0 1 ml of serum The precipitate was separated by centrifugation and the radioactive supernatant fluid was poured off Determination of the quantity of total radioactivity was obtained by adding that contained in the TCA precipitate to that in the TCA soluble fractions The quantity of total circulating radioactivity was also checked by placing 0 1 ml of serum on Whatman No 3 filter paper as in the procedure for electrophoresis However, in this instance the paper was placed immediately in 3 cc KOH Radioactivity in all samples was counted in a shielded well type scintillation crystal connected through a linear preamplifier to a scaler The gamma emissions of the I^{131} were measured to eliminate errors of interference All counts were carried out to sufficient numbers to reduce the counting error to less than 2 per cent

TABLE 1
PERCENTAGES OF DEVIATION FROM THE MEAN OF THREE CONSECUTIVE SERUM ANALYSES

Compartments	Deviation from mean			Average
Total radioactivity	+1	−3	+1	±1 4
TCA precipitate	+1	0	−2	±1 0
TCA soluble	+1	−3	+1	±1 4
Insulin	−5	+2	+1	±2 6
Average error per cent total count				±1 6

Determinations of the radioactivity in the TCA precipitable and the TCA soluble insulin and of the total compartments were carried out in duplicate on both the serum of the rabbit and the serum phantom Three consecutive serum phantoms were compared for accuracy The deviations of determinations of each of the serum compartments from their mean are shown in TABLE 1 The average error introduced by each analysis was 1 6 per cent of the total counts

Results

It has been demonstrated that insulin labeled with I^{131} can be separated from radioactive proteins in serum more accurately by electrophoresis than by precipitation with trichloroacetic acid The assumption that all radioactivity precipitated from the serum by trichloroacetic acid is biologically active insulin was not supported by previous work from this laboratory [1] The importance of using a technique with which the compartments can be

clearly distinguished is shown in FIGURE 1. The first column shows the characteristics of the radioactive material assayed in serum immediately after its arrival from the manufacturer. Ninety two per cent of the radioactivity in solution can be precipitated by trichloroacetic acid. The remaining 8 per cent is in the TCA soluble fraction. Only 86 per cent of the total radioactivity can be isolated as insulin by electrophoresis. Subtraction of the quantity of radioactivity isolated as insulin by electrophoresis from the TCA precipitable fraction leaves the radioactive fraction due to biologically inactive material. Columns 2 and 3 show the characteristics of the material at 4 and 8 days, respectively. The insulin fraction is now 77 and 74 per cent of total radioactivity, the inactive fraction has risen from 4 to 15 and 17

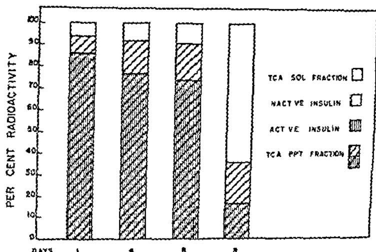


FIGURE 1. Characteristics of the radioactive solution. Radioactive solution stored at 10° C was added to serum on the days shown. Analysis was carried out immediately following mixing. Composition of the radioactivity in solution changes gradually with time. The fourth column shows the change in the characteristics of the solution kept at room temperature over an 8-day period.

per cent respectively over this period. The fourth column shows the characteristics of a part of the material that was separated from the original but left at room temperature. The insulin content is reduced to 17 per cent of the total radioactivity after 8 days; the TCA soluble fraction is 64 per cent of the total radioactivity. It is important to note that the characteristics of the radioactive solution may change completely. Unless the characteristics of the radioactive solution are followed, reduction in the insulin content as shown here will be undetected. This will alter the results obtained and studies done at different times will not be comparable.

The biologically inactive material migrates with the serum protein on electrophoresis of the serum. There is no detectable degradation of this fraction over a period of several hours and it continues to recirculate in the plasma.[1] Therefore the TCA soluble radioactivity that appears in the

plasma or serum following injection of the radioactive solution comes from the degradation of the insulin. These products of degradation will hereafter be designated as soluble I^{131}.

The rate of insulin destruction *in vivo* may be determined by the following technique. The amount of radioactivity injected as insulin I^{131} and the soluble I^{131} are measured by analyses of the serum phantom. At the end of the experimental period blood is drawn for determination of the serum concentration of insulin and soluble I^{131}. The bladder is removed from the intact animals for determination of soluble I^{131} that has been excreted in the urine. The soluble I^{131} resulting from insulin degradation that is not

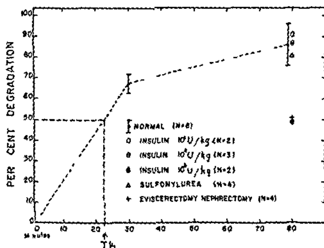


FIGURE 2. Insulin degradation in the rabbit. Degradation of radioinsulin injected in the rat but has been studied under various conditions. Prior administration of insulin in dosages of 10^1 and 10^2 U/kg did not alter the degree of degradation. Sulfonylurea (tolbutamide, chlorpropamide) had no effect on degradation. Insulin in dosage of 10^3 U/kg in the intact animal decreased degradation to the level of that in the eviscerated nephrectomized animal.

excreted is calculated by multiplying the I^{131} content of the serum by the iodine space. The iodine space was found to be 26 per cent of body weight and is in agreement with previous determinations.[4] The intact insulin remaining at the end of the experiment is the difference between the injected radioactivity and the total degradation products.

The per cent of insulin degradation at 30 min and 80 min in the normal rabbit is shown in FIGURE 2. At 30 min after injection an average of 68 per cent of injected radioinsulin has undergone separation of the radioactive label. This is assumed to be the result of proteolytic destruction of the insulin molecule. The half time of insulin destruction is 22 min. This correlates closely with the half time of the disappearance of radioactivity from the plasma as previously reported.[1] At 80 min an average of 86

per cent of the injected radioinsulin has been degraded This may be contrasted to the rate of degradation of insulin in the eviscerated nephrectomized rabbit As shown in FIGURE 2 the degradation of insulin was decreased to 51 per cent at 80 min under these circumstances

In order to slow the process of degradation in the normal rabbit, an attempt was made to saturate the degrading mechanisms with crystalline zinc insulin Dosages of crystalline zinc insulin of 1 10 100 and 1000 U/kg body weight were injected 10 min prior to the injection of radioinsulin The rate of insulin degradation was not influenced by quantities of insulin up to 100 U/kg body weight When 1000 U of insulin/kg body weight were injected prior to the radioinsulin the rate of degradation was reduced to 48 per cent at

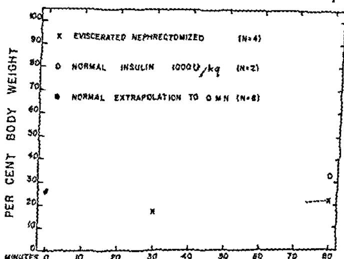


FIGURE 3 The apparent volume of distribution of insulin in the rabbit is equivalent to the volume of the extracellular space in the insulin treated (10^3 U/kg) and eviscerated nephrectomized rabbit Previous determinations in the normal animal[3] are also shown for comparison

80 min It is clear that the degrading mechanisms for insulin are difficult to saturate and far exceed their physiological need

The possibility that the sulfonylurea compounds act by decreasing insulin destruction is suggested by some of the studies done *in vitro* with enzyme systems[6] On the other hand no change in the rate of disappearance of radioinsulin following tolbutamide administration was noted[7] The effect of tolbutamide and chlorpropamide on insulin degradation has been studied with the present technique As shown in FIGURE 2 there was no decrease in the rate of insulin destruction following the use of these drugs in the normal rabbit

In order to clarify the role of tissue binding in the disappearance of insulin the space of distribution of insulin must also be determined It is probable that tissue binding and the space of distribution are closely related A determination of insulin space was made in earlier studies from this

laboratory in which it was assumed that the rate of degradation was constant from the time of injection Following injection of radioinsulin determinations of the insulin content of serum were made between 30 and 80 min after injection The rate of insulin disappearance was extrapolated to zero time The insulin content of serum at zero time was divided into the quantity of insulin given by injection By this calculation the insulin space was found to be approximately 26 per cent of total body weight[1] The insulin space was not altered by prior administration of sulfonylurea[7]

The present technique has been applied also to the study of the insulin space The quantity of intact insulin remaining at 30 or 80 min divided by the insulin concentration per ml of serum at the same intervals gives the apparent volume of distribution of the intact insulin Insulin destruction by the liver and kidney was eliminated in these experiments in order to clarify the effect of the peripheral tissue on insulin disappearance As shown in FIGURE 3 the insulin space was found to be 16 per cent of body weight at 30 min and 22 per cent of body weight at 80 min in the eviscerated nephrectomized rabbit Also shown are the results obtained when degradation is decreased by the prior intravenous administration of insulin in a dosage of 1000 U kg The results in FIGURE 3 are in accordance with the previous estimations of the insulin space[1 8] and are approximately the size of the extracellular space

The implications of this apparent volume of distribution of insulin should be considered further The quantity of insulin necessary to saturate the binding sites for insulin in muscle has been estimated by Stadie[9] to be approximately 1 5 µg of insulin gm of tissue Under the conditions of the experiments reported here the availability of tissue binding sites must greatly exceed the available insulin If intact insulin were captured by peripheral cells without degradation the quantity of insulin remaining in the periphery would increase while the concentration of circulating insulin would become progressively smaller Therefore the apparent volume of distribution of the intact insulin would become progressively larger and finally become indeterminate The apparent volume of distribution found in these experiments is inconsistent with this assumption The radioactive insulin injected can be accounted for by the summation of the insulin subjected to proteolytic destruction and the insulin contained in extracellular water This leads to the interpretation that the degradation of intact insulin is not prevented by tissue binding or sequestration The similarity between the space occupied by intact insulin and the extracellular compartment suggests rather that insulin is degraded quickly after leaving the extracellular space The clinical implication of these results is that intact insulin cannot be saved for future use by peripheral tissues

Summary

Factors influencing the rate of disappearance of insulin from plasma have been studied by the use of insulin labeled with iodine 131 (I^{131}) A technique has been employed that measures the amount of active intact insulin administered Some change in the characteristics of the radioactive solution on

standing for short periods makes this necessary In the intact animal an average of 86 per cent of the injected insulin was degraded in 80 min The rate of degradation was not influenced by prior administration of sulfonyl urea compounds

The apparent volume of distribution of insulin *in vivo* is equivalent to the volume of extracellular water Sequestration of intact insulin without degradation is not a factor in the rapid disappearance of insulin from plasma The radioactive insulin injected can be accounted for by the summation of the insulin subjected to proteolytic destruction and the insulin contained in extra cellular water Under the conditions of these experiments, there is no evidence that the degradation of intact insulin is prevented by tissue binding or sequestration

Acknowledgments

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A COMPARISON OF THE INFLUENCE OF TOLBUTAMIDE AND SMALL DOSES OF INSULIN ON THE SPLANCHNIC OUTPUT AND PERIPHERAL UPTAKE OF GLUCOSE IN MAN*

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Considerable evidence has been offered to support the hypothesis that the administration of a sulfonylurea to a human subject or animal with an intact pancreas is followed by the release of a substance with hypoglycemic activity, presumably insulin, from the pancreatic beta cells.[1] The refractoriness of fully alloxanized animals and the influence of the beta cell reserve on the character of the early hypoglycemic response to the drugs[2] are compatible with this hypothesis. In addition, pancreatic vein blood from dogs given a sulfonylurea reduced the blood glucose concentration of both normal[3] and alloxan-diabetic[4] recipient dogs in cross circulation experiments and produced hypoglycemia in mice.[5]

The unqualified acceptance of insulin as the mediator of sulfonylurea induced hypoglycemia has been tempered by the failure of many investigators to demonstrate an insulinlike effect of the drugs on the peripheral tissues.[6] In addition, the sulfonylureas have been shown to reduce the rate of glucose release from the liver,[7] while the existence of a direct effect of insulin on hepatic glucose metabolism has been questioned.[8]

Although an increase in the peripheral arteriovenous (A-V) blood glucose concentration difference has usually been observed in human subjects following the intravenous administration of a single dose of 0.1 U. insulin/kg or a total of 5 to 7 U.,[9, 10] doses of 3 U. or less have frequently failed to produce [illegible]

body glucose pool and suppressed the introduction of newly released glucose into the pool (glucogenesis), presumably from the liver. When insulin was given by the subcutaneous route in order to retard its rate of entrance into the circulating blood, 10 U. decreased the rate of glucogenesis without affecting glucose removal. A reduced net splanchnic output of glucose (NSGO) has been observed in man after either tolbutamide[12-14] or insulin.[15] These findings of various investigators suggested that in man small doses of insulin

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† Markle Scholar in The Medical Sciences.

might have the same effects as the sulfonylureas on the NSGO and the peripheral uptake of glucose

This possibility has been tested by comparing the effects of tolbutamide and small doses of insulin on the NSGO and the simultaneously measured peripheral A V blood glucose concentration difference in man

Methods

The subjects for these studies were 25 nondiabetic hospital patients who were tested after an overnight fast Eight of the subjects were tested with sodium tolbutamide * 11 received glucagon free crystalline insulin,† the other 6 served as control subjects and were given only 0 9 per cent sodium chloride solution These substances were administered into a peripheral vein during a 2 min interval in the following doses 2 gm of sodium tol butamide either 2 or 3 U of insulin and 50 ml of sodium chloride solution Blood samples were obtained simultaneously through indwelling needles in a radial artery and an antecubital vein and through a radiopaque catheter with its tip in a hepatic vein Simultaneous arterial, peripheral venous and hepatic venous blood samples were obtained at 5 or 10 min intervals during a 15 to 35 min preliminary control period and for 50 min after injection of the test substance The glucose content of the blood samples was determined in duplicate by the titrimetric method of Somogyi,[16] by this method a total of 23 determinations on 4 blood samples yielded a mean value of 85 mg 100 ml with a mean standard deviation of 2 1 mg /100 ml The hepatic blood flow was estimated by the Bromsulphalein technique of Bradley *et al* [17] with measurement of the plasma Bromsulphalein concentra tion [illegible]

the [illegible]

and [illegible]

net output of glucose from the splanchnic system and a negative value would indicate a net uptake The statistical significance of the differences between mean values was assessed by the t test of Fischer

Results

As seen in FIGURE 1, tolbutamide produced an average maximum arterial [illegible] 100 ml in 40 min that was still maintained at [illegible] milar magnitude, with an [illegible] min and a prompt return [illegible] who were given sodium chloride no significant alteration in arterial blood glucose concentration was observed

The average values for the HV A blood glucose concentration difference are illustrated in FIGURE 2 and are summarized in TABLE 1 After tolbutamide administration the HV A difference decreased in all 8 subjects In 7 of the 8 subjects the average HV A difference during the first 30 min after tolbut amide was at least 4 mg /100 ml lower than the average preliminary control

* Supplied by C J O Donovan of The Upjohn Company Kalamazoo Mich
† Supplied by W R Kirtley of Eli Lilly and Company Indianapolis Ind

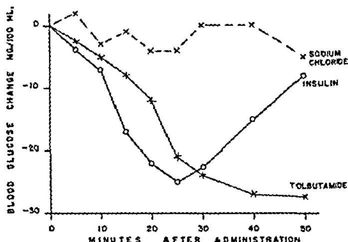


FIGURE 1 Average changes in arterial blood glucose concentration produced by the peripheral intravenous administration of sodium tolbutamide (8 subjects), insulin (9 subjects), or sodium chloride (6 subjects)

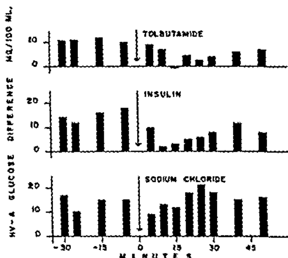


FIGURE 2 Average HV-A blood glucose concentration differences before and after the peripheral intravenous administration of sodium tolbutamide (8 subjects), insulin (9 subjects) or sodium chloride (6 subjects) The first 4 bars on the left in each section represent values obtained during a preliminary control period

TABLE 1

AVERAGE VALUES (±1 STANDARD DEVIATION) OF HV-A BLOOD GLUCOSE DIFFERENCE, NSGO, PERIPHERAL A-V BLOOD GLUCOSE DIFFERENCE AND (A-V)/A RATIO IN NONDIABETIC SUBJECTS BEFORE AND FOR THE FIRST 30 MIN AFTER THE PERIPHERAL INTRAVENOUS ADMINISTRATION OF SODIUM TOLBUTAMIDE, INSULIN OR SODIUM CHLORIDE

Substance administered	No. of subjects	Before	After	p
		HV-A (mg/100 ml)		
Sodium tolbutamide	8	10.7 ± 6.5	4.1 ± 3.2	<0.05
Insulin	9	14.9 ± 7.6	5.7 ± 3.0	<0.01
Sodium chloride	6	14.1 ± 5.8	15.3 ± 10.4	>0.5
		NSGO (mg/min)		
Sodium tolbutamide	8	138 ± 59	50 ± 29	<0.01
Insulin	9	197 ± 101	73 ± 25	<0.01
Sodium chloride	6	176 ± 69	199 ± 120	>0.5
		A-V (mg/100 ml)		
Sodium tolbutamide	6	2.2 ± 2.5	−1.0 ± 2.4	0.05
Insulin	9	2.9 ± 4.7	1.4 ± 3.8	>0.4
Sodium chloride	4	2.0 ± 1.7	3.0 ± 1.9	>0.4
		(A-V)/A ratio		
Sodium tolbutamide	6	.030 ± .046	−.021 ± .036	>0.05
Insulin	9	.028 ± .044	.019 ± .050	>0.5
Sodium chloride	4	.023 ± .023	.032 ± .025	>0.5

value Similarly after insulin treatment there was a diminution in the HV A difference in 9 of the 11 subjects In 8 of these 9 subjects the average decrease exceeded 4 mg/100 ml * After sodium chloride none of the control subjects showed a decline in the average HV A difference of as much as 4 mg/100 ml and there was no significant change in this difference for the control group as a whole

The average values for the NSGO are illustrated in FIGURE 3 and are summarized in TABLE 1 Tolbutamide decreased the NSGO in all 8 subjects The average value during the first 30 min after tolbutamide was at least 40 mg min less than the preliminary control value in all 8 subjects NSGO

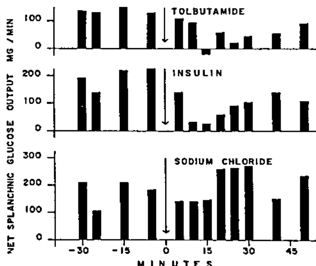


FIGURE 3 Average values for the NSGO before and after the peripheral intravenous administration of sodium tolbutamide (8 subjects) insulin (9 subjects) or sodium chloride (6 subjects) The first 4 bars on the left in each section represent values obtained during a preliminary control period

decreased after insulin in 9 of the 11 subjects tested in 8 of the 9 the decrease in the average NSGO exceeded 40 mg min After sodium chloride only

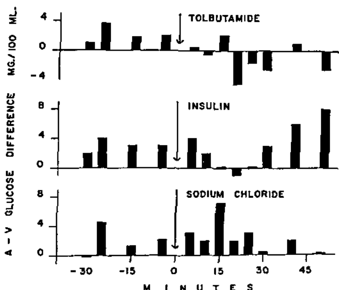
Although there were fluctuations in the estimated hepatic blood flow of individual subjects none of the substances administered produced a significant change in the average flow

The average values for the peripheral AV blood glucose concentration difference are illustrated in FIGURE 4 and are summarized in TABLE 1 No apparent increase in the AV difference was observed during the first 30 min

* Data from the two subjects in whom there was no demonstrable decrease in the HV A difference after insulin administration are not included in the figures or table

after tolbutamide in 5 of the 6 subjects tested In 3 of these 5 cases the average A V difference decreased more than 4 mg /100 ml and in 4 of them the values became negative During the first 30 min after insulin there was no increase in the average A V difference in 6 of the 9 subjects, the increase

glucose toward the initial fasting level at 40 and 50 min after insulin administration was accompanied by an increased peripheral A V difference that was



... before

section represent values obtained during a preliminary control period

at least 4 mg / 100 ml greater than the preliminary control value in 5 of the 9 subjects None of the 4 control subjects tested with sodium chloride showed an increase in A V difference as great as 4 mg /100 ml and there was no significant change in the A V difference for the group as a whole

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that was accompanied by a decrease in NSGO without an increase in peripheral *A V blood glucose concentration difference demonstrable by the* techniques employed

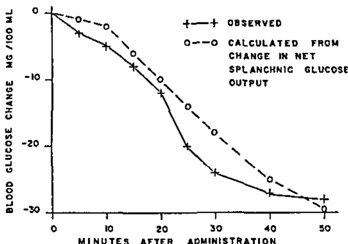


FIGURE 5 A comparison of the average arterial blood glucose concentration changes after tolbutamide administration with the calculated changes that would be produced by the observed alterations in NSGO The assumptions involved in this calculation are discussed in the text

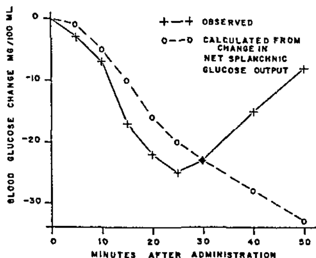


FIGURE 6 A comparison of the average arterial blood glucose concentration changes after insulin administration with the calculated changes that would be produced by the observed alterations in NSGO The assumptions involved in this calculation are discussed in the text

Discussion

The techniques employed in this study impose certain limitations on the conclusions that may be drawn from the results obtained

(1) Since the A-V glucose differences were measured only across the splanchnic bed and in an upper extremity changes in glucose assimilation in other organs would not be detected

(2) A decrease in NSGO could result from a diminution in the net hepatic glucose output an increase in the glucose uptake in an extrahepatic splanchnic component such as adipose tissue or both The present results do not allow a distinction among these possibilities and do not permit a decision as to whether tolbutamide and insulin act at the same or different sites within the splanchnic system Studies with C^{14} labeled glucose in animals[7, 20, 21] and man[13, 27] have shown that sulfonylurea induced hypoglycemia is accompanied by an inhibition of glucogenesis presumably from the liver From similar studies with insulin some investigators[13, 20, 2, 5] have concluded that one action of the hormone was a reduction of the rate of glucogenesis while others[7, 1] have been unable to demonstrate such an effect consistently Direct measurements of the hepatic venous portal venous glucose difference in the dog have shown a decrease in net hepatic glucose output after tolbutamide, but not after insulin administration[7] Such direct measurements are not available in man

(3) The limitations of the analytical method for glucose and the necessity for assuming the constancy of the peripheral blood flow make the measurement of the A-V glucose difference a relatively insensitive technique for the detection of small changes in peripheral glucose uptake The failure to detect an increment in A-V glucose difference by this technique does not exclude the occurrence of small increases in this difference and therefore should not be considered to prove that neither tolbutamide nor insulin in this dosage exerts any peripheral effect On the other hand if changes as large as those observed in the splanchnic system occurred in skeletal muscle and skin there is no reason to believe that they would escape detection since the total blood flow to resting skeletal muscle and skin in man is similar in magnitude to the splanchnic blood flow[26, 7]

(4) The present studies do not indicate the mechanism of action of tolbutamide and insulin The two hypoglycemic agents might have different and independent mechanisms of action even though their sites of action were the same

(5) In man the sites of action of endogenous and exogenous insulin might differ Such a discrepancy might be due to differences *in their loci of entry* into the blood stream[4] alterations produced in the exogenous hormone during its preparation[29] or the *heterologous nature of exogenous insulin* from animal sources[30]

It is of interest to determine whether the *observed decrease in NSGO* is adequate to account for the fall in the arterial blood glucose concentration that followed tolbutamide or insulin *administration* *If it is* assumed that the rate of glucose uptake by the nonsplanchnic tissues remained constant

and that glucose was distributed uniformly in the extracellular fluid volume, estimated as 20 per cent of the body weight,[31] the decline in the blood glucose concentration that would result from the observed decrease in NSGO can be calculated. The results of these calculations are shown in FIGURES 5 and 6. Although there are large discrepancies between the predicted and observed blood glucose decreases in individual cases, the average values are similar in magnitude after both tolbutamide and insulin. It is concluded that the decrease in NSGO could be the major cause of the blood glucose fall after tolbutamide or insulin, but the occurrence of an increased glucose uptake in a nonsplanchnic tissue cannot be excluded. In these studies the return of the blood glucose concentration toward the initial fasting level after insulin induced hypoglycemia is not adequately accounted for by the observed changes in NSGO or peripheral A V glucose difference.

The similarity of the results obtained with tolbutamide and small doses of insulin appears compatible with an insulin-mediated hypoglycemic action of the sulfonylureas. However, because of the limitations of the techniques employed, other mechanisms of action are not excluded.

Summary

In nondiabetic human subjects the arterial blood glucose decline produced by the peripheral intravenous administration of tolbutamide was accompanied by a reduction in NSGO sufficient to account for the major portion of the blood glucose fall. No increase in the peripheral A V glucose difference was demonstrated.

Similar changes were produced by small doses of insulin.

The similarity of the effects produced by tolbutamide and insulin is compatible with an insulin-mediated hypoglycemic action of the sulfonylureas. This conclusion must be tempered by recognition of the limitations of the experimental techniques employed.

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THE RELATIONSHIP BETWEEN THE MECHANISM OF ACTION OF THE SULFONYLUREAS AND THE SECRETION OF INSULIN INTO THE PORTAL CIRCULATION*

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The conclusion that the hypoglycemic effect of the sulfonylureas is mediated through the secretion of endogenous insulin is based upon data indicating that the presence of functional pancreatic beta cells is prerequisite for their hypoglycemic action. In all experimental preparations in which the beta cells are ablated, including completely alloxanized, depancreatized, or eviscerated animals, the sulfonylureas do not significantly lower the blood sugar.[1-4] By contrast, hypoglycemia eventuates whenever the beta cells are intact,[5] even in hepatectomized animals.[6,7] Recently Pfeiffer has reported an increase in insulinlike activity of the portal venous blood after sulfonylurea administration.[8]

Despite the formidable evidence linking the hypoglycemic effect with the presence of beta cells, objections to the insulinogenic mechanism of action have been raised. For the most part these objections are based upon the failure of the sulfonylureas to duplicate precisely the metabolic effects of exogenous insulin on a variety of parameters. The sulfonylureas are said to produce hypoglycemia by decreasing the hepatic output of glucose.[9-12] This hepatic effect and the failure of the sulfonylureas to increase peripheral glucose utilization consistently[13,14] have been cited as physiological actions inconsistent with the beta-cytotropic mechanism of action. It should be pointed out that such objections implicitly assume that endogenous insulin reduces blood glucose concentration primarily by increasing peripheral glucose utilization and has little or no effect on the hepatic output of glucose.

To conclude from these conflicting data that the sulfonylureas do not stimulate endogenous insulin secretion leaves their dependence on intact beta cells obscure and unexplained. In view of this beta cell dependence, an alternative conclusion would be that the sulfonylureas stimulate the secretion of endogenous insulin which, in turn, may differ in its physiological effects from exogenous insulin. The natural hormone and its commercial counterpart, exogenous insulin, may differ for several reasons: (1) exogenous insulin may be altered in its commercial preparation, (2) exogenous insulin prepared from one species and administered to another species may not have physiological effects identical to the species specific endogenous hormone, (3) the metabolic effect of exogenous insulin may be altered because it enters the body through the systemic circulation. Under physiological conditions endogenous insulin is secreted into branches of the portal circulation, there

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† Established Investigator, American Heart Association, Inc., New York, N. Y.

fore, *all* of the natural hormone must traverse the liver before entry into the systemic circulation, whence it becomes available to the peripheral tissues

The delivery of endogenous insulin to the liver in amounts and concentration greater than those possible for any other tissue in the body suggests that the secretion into the portal circulation has physiological significance Nevertheless the controversy as to whether insulin has a direct effect on the hepatic output of glucose still persists [11, 15, 16] *In vitro* effects of insulin on the liver have been difficult to demonstrate but *in vivo* effects of varying duration and magnitude have been reported [17, 19] Recently new evidence both *in vitro*[19] and *in vivo* [20, 23] supporting a direct effect of insulin on hepatic glucose metabolism has appeared The data of Wall [20] Dunn [21] Jacobs [23] and their colleagues obtained by the technique of determining changes in the specific

intravenously there results only a transient decrease in hepatic glucose output and a prolonged increase in peripheral glucose utilization of considerable magnitude By contrast when insulin is administered subcutaneously or by slow intravenous infusion a depression of hepatic glucose output of considerable duration ensues In the present studies different parameters the arteriovenous (A V) glucose difference across the leg[24] and the hepatic output of gl co e[25]

subjects during the intravenous infusion of glucagon free insulin administered for 120 min at a rate of 0 06 to 0 1 U min The changes in A V glucose difference were then compared to changes that occurred in the same 8 subjects following the intravenous administration of 1 gm of sodium tolbutamide Femoral arterial and venous blood samples were collected simultaneously at 10 min intervals during a 150- to 180 min period by means of indwelling Cournand needles Blood glucose concentrations were determined by the Somogyi copper iodometric method[26] on 2 ml blood samples

During insulin infusion mean arterial blood glucose concentration fell from 80 to 52 mg per cent femoral A V glucose difference failed to increase and averaged only 1 8 mg per cent Similar results occurred after tolbutamide administration Mean blood glucose concentration fell from 83 to 52 mg per cent and A V glucose differences averaged 2 3 mg per cent The data indicate no significant difference in peripheral glucose utilization as manifest by changes in A V glucose difference after tolbutamide and after the slow infusion of insulin While it is possible that peripheral glucose utilization was increased but could not be detected after insulin administration, these data can be alternately interpreted to indicate an hepatic effect of both tolbutamide and insulin under these experimental conditions Such an interpretation is in accord with the previously mentioned data of Jacobs *et al* who noted an hepatic effect of insulin administered subcutaneously or by slow intravenous infusion [22] A typical set of experiments showing the effects on A V glucose difference of insulin and tolbutamide are shown in FIGURES 1 and 2

THE RELATIONSHIP BETWEEN THE MECHANISM OF ACTION OF THE SULFONYLUREAS AND THE SECRETION OF INSULIN INTO THE PORTAL CIRCULATION*

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The conclusion that the hypoglycemic effect of the sulfonylureas is mediated through the secretion of endogenous insulin is based upon data indicating that the presence of functional pancreatic beta cells is prerequisite for their hypoglycemic action In all experimental preparations in which the beta cells are ablated including completely alloxanized depancreatized or eviscerated animals the sulfonylureas do not significantly lower the blood sugar [1-4] By contrast hypoglycemia eventuates whenever the beta cells are intact [5] even in hepatectomized animals [6,7] Recently Pfeiffer has reported an increase in insulinlike activity of the portal venous blood after sulfonyl urea administration [8]

Despite the formidable evidence linking the hypoglycemic effect with the presence of beta cells objections to the insulinogenic mechanism of action have been raised For the most part these objections are based upon the failure of the sulfonylureas to duplicate precisely the metabolic effects of *exogenous insulin on a variety of parameters The sulfonylureas are said to* produce hypoglycemia by decreasing the hepatic output of glucose [9-12] This hepatic effect and the failure of the sulfonylureas to increase peripheral glucose utilization consistently[13,14] have been cited as physiological actions inconsistent with the beta-cytotropic mechanism of action It should be pointed out that such objections implicitly assume that endogenous insulin reduces blood glucose concentration primarily by increasing peripheral glucose utilization and has little or no effect on the hepatic output of glucose

To conclude from these conflicting data that the sulfonylureas do not stimulate endogenous insulin secretion leaves their dependence on intact beta cells obscure and unexplained In view of this beta-cell dependence, an alternative conclusion would be that the sulfonylureas stimulate the
physiological
ts commercial
(1) exogenous
insulin may be altered in its commercial preparation (2) exogenous insulin

the body through the systemic circulation Under physiological conditions endogenous insulin is secreted into branches of the portal circulation, there

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fore *all* of the natural hormone must traverse the liver before entry into the systemic circulation whence it becomes available to the peripheral tissues

The delivery of endogenous insulin to the liver in amounts and concentration greater than those possible for any other tissue in the body suggests that the secretion into the portal circulation has physiological significance Nevertheless the controversy as to whether insulin has a direct effect on the hepatic output of glucose still persists [11, 15, 16] *In vitro* effects of insulin on the liver have been difficult to demonstrate but *in vivo* effects of varying duration and magnitude have been reported [17, 18] Recently new evidence both *in vitro*[19] and *in vivo* [20-23] supporting a direct effect of insulin on hepatic glucose metabolism has appeared The data of Wall [20] Dunn [21] Jacobs [23] and their colleagues obtained by the technique of determining changes in the specific activity of a trace dose of uniformly labeled glucose C^{14} after insulin administration indicate that the rate of insulin administration appears to affect the magnitude of the hepatic response When insulin in large amounts is injected intravenously there results only a transient decrease in hepatic glucose output and a prolonged increase in peripheral glucose utilization of considerable magnitude By contrast when insulin is administered subcutaneously or by slow intravenous infusion a depression of hepatic glucose output of considerable duration ensues In the present studies different parameters the arteriovenous (A V) glucose difference across the leg[24] and the hepatic output of glucose [25] were measured during the slow infusion of glucagon free insulin and a similar depression of hepatic glucose output was found

Changes in femoral A V glucose difference were determined in 8 normal subjects during the intravenous infusion of glucagon free insulin administered for 120 min at a rate of 0 06 to 0 1 U min The changes in A V glucose difference were then compared to changes that occurred in the same 8 subjects following the intravenous administration of 1 gm of sodium tolbutamide Femoral arterial and venous blood samples were collected simultaneously at 10 min intervals during a 150- to 180-min period by means of indwelling Cournand needles Blood glucose concentrations were determined by the Somogyi copper iodometric method[26] on 2 ml blood samples

During insulin infusion mean arterial blood glucose concentration fell from 80 to 52 mg per cent femoral A V glucose difference failed to increase and averaged only 1 8 mg per cent Similar results occurred after tolbutamide administration Mean blood glucose concentration fell from 83 to 52 mg *per cent and A V glucose differences averaged 2 3 mg per cent* The data indicate no significant difference in peripheral glucose utilization as manifest by changes in A V glucose difference after tolbutamide and after the slow infusion of insulin While it is possible that peripheral glucose utilization was increased but could not be detected after insulin administration these data can be alternately interpreted to indicate an hepatic effect of both tolbutamide and insulin under these experimental conditions Such an interpretation is in accord with the previously mentioned data of Jacobs *et al* who noted an hepatic effect of insulin administered subcutaneously or by slow intravenous infusion [23] A typical set of experiments showing the effects on A V glucose difference of insulin and tolbutamide are shown in FIGURES 1 and 2

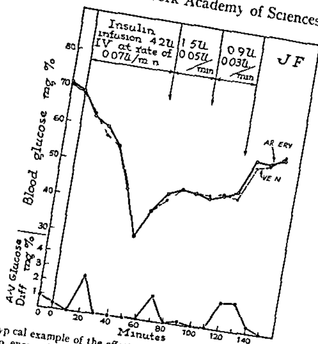


FIGURE 1. Typical example of the effects of the slow infusion of glucagon free insulin on femoral arterio-venous glucose difference in normal subjects.

FIGURE 2. Typical example of the effects of the intravenous administration of 1 gm of sodium tolbutamide on the femoral arterio-venous glucose difference in a normal

In order to determine by direct measurement whether the slow infusion of insulin results in a decreased hepatic output of glucose hepatic blood flow and hepatic vein arterial glucose differences were measured in normal and diabetic subjects Following hepatic vein catheterization hepatic blood flow was measured by the Bromsulphalein technique of Bradley *et al*[27] Simultaneous hepatic venous and arterial blood samples were collected at 10-min intervals for 20 to 30 min prior to and during the 60 min duration of the insulin infusion Examples of two experiments one in a diabetic subject and the other in a normal subject are shown in TABLE 1 and in FIGURE 3 In both instances the hypoglycemia following the insulin infusion was associated with a profound reduction in the hepatic output of glucose

TABLE 1
EFFECT OF INSULIN INFUSION ON HEPATIC GLUCOSE OUTPUT IN A LABILE DIABETIC

Time (minutes)	Estimated hepatic blood flow (ml min)	Blood glucose (mg 100 ml)			Hepatic glucose output (mg min)
		Hepatic vein	Femoral artery	HV Art	
−25	962	450	270	180	1730
−15	1144	475	269	206	2360
− 5	1177	427	274	153	1800
Start insulin infusion 0 1 U min					
10	1030	373	275	98	1010
20	1121	395	258	137	1540
30	970	345	250	95	922
40	923	301	230	71	660
50	907	311	221	90	820
60	895	210	207	3	27
70	883	210	185	25	222

In order to obviate the criticism that these changes reflect only alterations in net splanchnic glucose output and not necessarily in net hepatic glucose output eight experiments were performed on dogs with complete portacaval shunts In this preparation all the blood perfusing the liver comes from the arterial circulation A typical example of the effect of the infusion of 0 025 U min of glucagon free insulin on hepatic venous and arterial blood glucose concentrations and on the hepatic output of glucose is shown in FIGURE 4

These data indicate that under proper experimental conditions exogenous insulin apparently has an effect upon hepatic glucose metabolism Since all endogenous insulin must traverse the liver before entering the systemic circulation the concentration and amount that reach the liver must exceed those reaching all other tissues This suggests that endogenous insulin

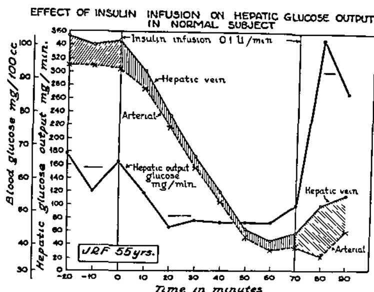


FIGURE 3 Effect of the slow infusion (0.1 U/min) of glucagon free insulin on the hepatic venous arterial glucose difference and estimated hepatic output of glucose in a normal subject

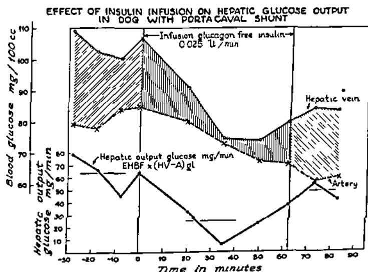


FIGURE 4 Effect of the slow infusion (0.025 U min) of glucagon free insulin on the hepatic venous arterial glucose difference and hepatic output of glucose in a dog with a complete portacaval shunt

may have a greater hepatic effect than insulin administered into the systemic circulation and therefore may account for the hepatic effect of the sulfonylureas

In order to determine whether the initial passage of insulin through the liver alters the magnitude of its hepatic and peripheral actions the effects of glucagon free insulin administered by the portal vein and by a peripheral vein were compared in 16 experiments on 8 dogs The details of these experiments have already been reported [28] In these studies the magnitude of the arterial hypoglycemia was the same after both routes of administration However in each dog the peripheral injection of insulin resulted in a significantly greater increase in peripheral glucose utilization than did the portal administration The combination of similar arterial hypoglycemia coupled with a lesser augmentation of peripheral glucose utilization after portal injection suggests that [illegible]

vein than when injected into the inferior vena cava [29] A typical example of the comparative effects of the portal and peripheral routes of insulin administration on the magnitude of arterial hypoglycemia and on the magnitude of increase in peripheral glucose utilization is shown in FIGURE 5

The apparently greater hepatic effect of insulin injected by the portal vein suggests that a substantial amount of this insulin is retained by the liver during the initial transhepatic passage Studies were designed to determine whether a significant amount of insulin is retained by the liver during a single circulation and also to compare the peak hepatic binding of insulin after portal and peripheral administration [30] In the first study 196 rats were injected with I^{131} labeled insulin In one half of the rats the insulin (0 002 units) was injected into the portal vein in the other one half into the femoral vein The rats were sacrificed at frequent intervals from 4 sec to 15 *min after insulin injection Immediately thereafter the liver* was perfused with 30 to 50 cc of modified Ringer s solution The liver kidneys and other tissues were then assayed for radioactivity in a well type gamma counter and the percentage of the administered dose bound in each organ was determined The results indicate that during the single transhepatic circulation following portal injection 51 per cent of the administered insulin was bound firmly to the rat liver By contrast peak hepatic binding after the peripheral route of administration was only 27 per cent of the administered dose

In order to determine whether the human liver binds a significant amount of insulin during a single transhepatic circulation the following study was performed in 12 human subjects without known hepatic or pancreatic disease [31] During abdominal operation a solution containing 0 [illegible] of I^{131} labeled insulin and a known amount of inulin was injected rapidly into the portal vein Ten seconds later a brachial arterial blood sample was collected over a 10 to 15 sec period The concentrations of inulin and insulin I^{131} were then determined Since known amounts of insulin I^{131} and inulin were

injected, their volumes of distribution could be calculated and compared. Control studies were performed in 8 subjects by injecting the solution of inulin and insulin I^{131} into the antecubital vein and collecting the arterial blood sample at precisely the same time as after portal injection.

In the control studies the volumes of distribution of labeled insulin (2058 ml) and inulin (1989 ml) were almost identical. By contrast, after portal injection the volume of distribution of the labeled insulin (5581 ml) was more than twice as large as that of inulin (2584 ml). This indicates that 52 per cent of the insulin was bound to the human liver in a single transhepatic circulation.

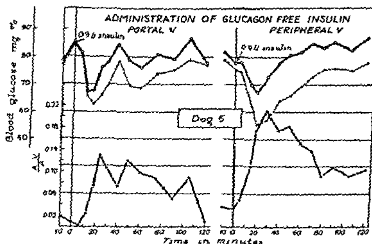


FIGURE 5. Typical example of the comparative effects of glucagon free insulin administered by the portal vein and by a peripheral vein on the magnitude of the hypoglycemia and the magnitude of the increase in peripheral arteriovenous glucose difference. Note that the degree of hypoglycemia is the same after both routes of administration and that the arteriovenous glucose differences are greater after the peripheral administration of insulin. Reproduced by permission of *The Journal of Clinical Investigation*.[29]

These data support but do not prove the concept that endogenous insulin in man regulates the hepatic output of glucose. However, they should not be construed to mean that this is the sole function of insulin. We have other data indicating that, when the hepatic insulin binding sites are saturated[32] or blocked,[33] a greater portion of endogenous insulin passes through the liver to the periphery where it exerts a greater effect on peripheral glucose utilization.[34] Crandall and Cherry have also previously reported that changes in the hepatic output of glucose follow anesthesia and acute operative stress.[35]

Summary and Conclusions

There is a growing body of evidence, not yet incontrovertible, indicating that insulin, when administered under proper experimental conditions, results in a decreased hepatic glucose output and thereby contributes to the

hypoglycemic effect The data from the present experiments support such an action Moreover, the data indicating that more than 50 per cent of labeled insulin injected intraportally is bound by the liver suggests that endogenous insulin may play a significant role in the regulation of the hepatic release of glucose

While these findings do not prove the beta cytotropic mechani m of action of the sulfonylureas, they are in complete accord with such a hypothesis The failure of the sulfonylureas to duplicate preci ely the effe ts of exogenous insulin may be related to the evidence presented here which indicates that the site rate, magnitude and duration of insulin administration apparently alter its subsequent metabolic effect For these reasons such differences cannot be used to controvert other formidable evidence in licating that the hypoglycemic action of the sulfonylureas is beta cell dependent and that these agents in rease the secretion of endogenous insulin

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CHANGES INDUCED BY INSULIN AND TOLBUTAMIDE IN THE GLUCOSE OUTPUT OF THE LIVER

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Following the independent observation of the additional hypoglycemic effect of the sulfonylureas in insufficiently insulin treated pancreatectomized dogs[1] it became evident that these compounds might produce the decrease in the blood sugar concentration of both normal and diabetic dog by inhibiting either partly or totally the release of glucose from the liver

The first part of this report describes our experimental observations after applying the single injection of C^{14} labeled glucose for measuring the glucose output in the liver in normal dogs before and after intravenous injections of either tolbutamide or insulin

A new method for calculating this output from the isotope data was developed By equaling the ratio between specific activity of glucose in samples of hepatic venous and arterial blood to the amount of glucose entering and leaving the liver, the output of unlabeled glucose from this organ could be calculated for any period before or after the administration of a substance producing a change in the size of the glucose pool This method may have an advantage over the ordinary isotope dilution method based upon injection of C^{14} glucose, which does not allow calculation of the glucose turnover during the actual changes of the plasma glucose concentration as the latter will not necessarily be representative of the various compartments of the glucose pool

The second part of the report deals with the results obtained with this method when insulin was infused portally into normal cyclopropane anesthetized dogs

*Effect of Intravenous Injections of Tolbutamide or Insulin**

These studies were performed in normal dogs fasted overnight and anesthetized with pentobarbital sodium (Nembutal) a dose of about 200 μc of uniformly labeled ... ected intravenously ... mples were collected e ... in a peripheral vein After a control period of about 1 hour a total dose of ... ly in 2 doses (0 10 gm kg ... mples were collected at ... As shown in FIGURE 1 the exponential decrease of the blood glucose specific activity observed in the control period, which reflected a constant release of unlabeled glucose from the liver to replace the amount of glucose utilized in the peripheral tissues was interrupted by the injection of the tolbutamide as the specific activity remained almost constant for a period of 30 to 40 min During this time the plasma glucose concentration decreased to about 40 to 50 mg per cent When these hypoglycemic levels were reached, the specific activity

again decreased exponentially at a rate slightly higher than in the control period but nevertheless *corresponding to* a lesser release of glucose from the liver

For the example shown in FIGURE 1 the rate of glucose release from the liver was about 57 mg /min in the control period Assuming that a similar

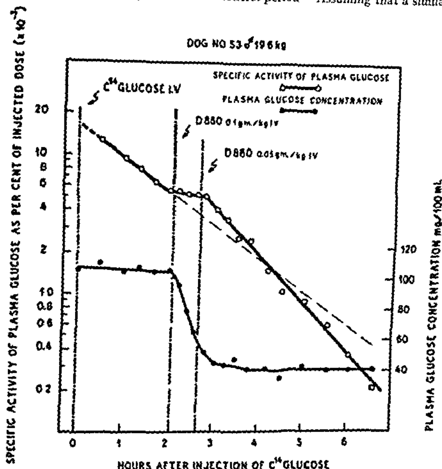


FIGURE 1 The effect of intravenously injected tolbutamide (D 860) on the plasma glucose concentration and specific activity in an anesthetized normal dog

rate of peripheral glucose utilization persists under the actual decrease in the blood sugar, this rate would be sufficient to decrease the blood sugar to the level observed, as this would require a utilization of about 65 mg /min, which is not significantly different from the control value The steeper decline later in the postinjection period, indicating the rate to be about 45 mg /min, does not reflect an increased rate of utilization, but is due to the considerably smaller glucose pool

Searle and Chaikoff[3] observed a similar plateau after creating a hyper

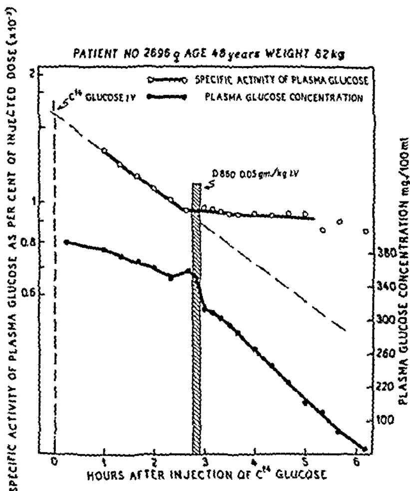


FIGURE 2 The effect of intravenously injected tolbutamide (D 860) on the plasma glucose concentration and specific activity in an unanesthetized diabetic patient who received the last injection of regular insulin 18 hours prior to the examination

glycemia that was interpreted as an inhibition of the glucose output Accord ing to this, the practically constant specific activity of the blood glucose after the injection of tolbutamide may also indicate a block in the glucose release from the liver The short duration of a complete inhibition is thought to be due to the measures taken by the organism to overcome the large degree of hypoglycemia shortly after it has been established

Actually this explanation was indirectly confirmed when a plateau in th specific activity curve was observed under identical experimental conditions

in 2 diabetic patients and 2 pancreatectomized dogs In these cases the plateau lasted for the entire postinjection period of observation (FIGURES 2 and 3) Although exogenous insulin (20 I U lente insulin) had been administered to the diabetic dogs only 18 hours before the injection of tolbutamide a very clear cut inhibition of the glucose output of the liver was produced (FIGURE 3) As a result of this the blood sugar decreased from about 550 mg per cent to about 250 mg per cent in the course of 4 hours This decrease was produced by the rate of glucose uptake in the peripheral tissues and the excretion in the urine

The experimental data of Creutzfeldt[4] and others indicate that the hypoglycemia might be produced by a stimulation of the release of insulin from the beta cells of the Langerhans islets Therefore identical experiments were carried out replacing tolbutamide with 2 intravenous injections of

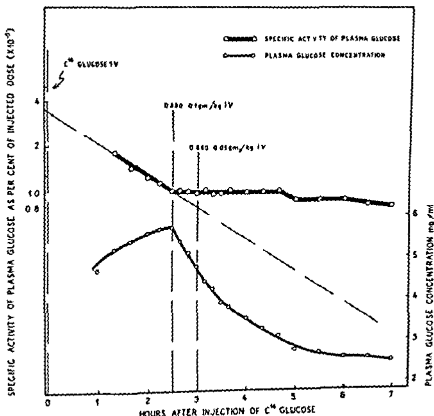


FIGURE 3 The effect of intravenously injected tolbutamide (D 860) on the plasma glucose concentration and specific activity in an anesthetized pancreatectomized dog which 18 hours before the injection of the C[14] glucose received its last dose of insulin (lente insulin 14 I U + insulin 6 I U)

in ulin (0 1 I U and 0 05 I U kg at 10 min intervals) *In contrast to the findings after tolbutamide* this was immediately followed by a considerably steeper decrease in the specific activity which cannot be produced by an inhibition of the glucose release from the liver (FIGURE 4)

In an independent study Dunn *et al* [6] observed a very short plateau (5 to 10 min) *in the specific activity curve following doses of insulin in the range*

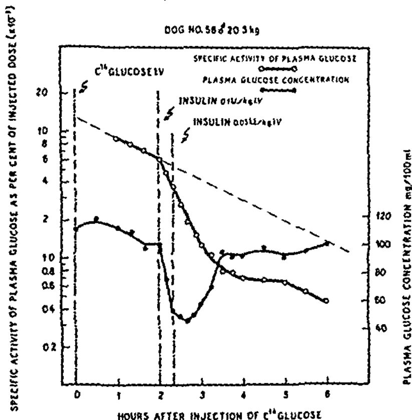


FIGURE 4 The effect of intravenously injected insulin on the plasma glucose concentration and specific activity in an anesthetized normal dog

of 1 I U kg *To a certain extent we can confirm this observation* as doses of insulin exceeding 0 5 I U kg produce what appears to be a very short lasting plateau indeed far shorter than that observed after tolbutamide *This may be related to these rather unphysiological doses* of insulin especially as it is not observed in our experiments when smaller and perhaps more physiological doses of insulin are injected Therefore from our point of view it is not likely that the hypoglycemic effect of tolbutamide is due to an enhancement of the insulin release from the pancreas

However, in order to test the possibility that an increase of the insulin secreted from the pancreas into the portal blood might have a special and limited effect upon the glucose output from the liver, the subsequent study of the effect of insulin constantly infused intraportally in amounts varying from 0 02 to 0 12 I U /kg /min was undertaken

The Glucose Output of the Liver Following Intraportal Insulin Infusions

The principles of the calculation of the amount of unlabeled glucose released from the liver are shown schematically in FIGURE 5 The specific activity

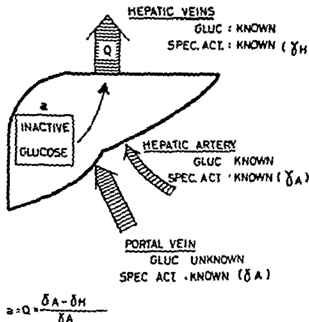


FIGURE 5 The principle of the isotope dilution method worked out for calculating the release of unlabeled glucose (a) from the liver (*Q* = total amount of glucose leaving the liver)

of the glucose in portal and arterial blood is identical if no dilution occurs with unlabeled glucose absorbed from the intestines This is confirmed experimentally in normal dogs starved for 18 hours, although the concentration of glucose may differ in blood from these two vessels The arterial specific activity can thus be regarded as representative for all blood glucose entering the liver If γA and γH represent the specific activity of glucose in arterial and hepatic venous blood, the ratio $\gamma A : \gamma H$ will be equal to the ratio between the amount of glucose leaving the liver (*Q*) and the amount passing through the liver (*D*) Hence

$$\frac{\gamma A}{\gamma H} = \frac{Q}{D} \qquad (1)$$

While D cannot be ascertained the value of Q is calculated from the estimated hepatic blood flow[8] and the hepatic venous glucose concentration, and further as

$$Q = D + a \tag{2}$$

where a is the amount of unlabeled glucose released from the liver, this amount can be derived as

$$a = Q\left(1 - \frac{\gamma H}{\gamma 1}\right) \tag{3}$$

This equation will be suitable for liver output calculations even in periods

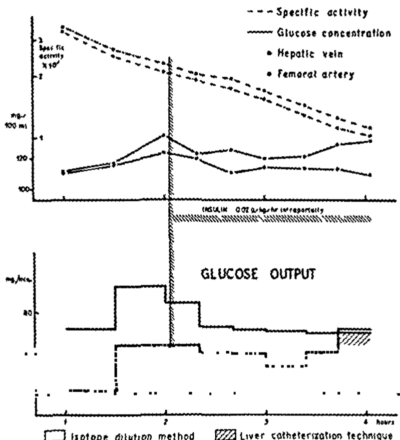


FIGURE 6 Effect of intraportal infused insulin (0.02 I.U. kg./hour) into a normal dog upon the output of unlabeled glucose from the liver and the net amount of glucose leaving the splanchnic area. In the upper part of the figure are shown the arterial and hepatic venous plasma glucose concentration and specific activity

where a steady state with respect to glucose metabolism is *not maintained* as variations in the glucose pool do not interfere

The output of unlabeled glucose from the liver before and during an intraportal infusion of 0.02 I.U. of insulin/kg./hour is shown in FIGURE 6 together with the net amount of plasma glucose leaving the splanchnic organs, calculated from the difference in glucose concentration of hepatic venous and arterial plasma multiplied by the estimated hepatic plasma flow. Although this dose of insulin does not have any apparent effect upon the arterial blood sugar concentration, it could be anticipated that if insulin effects the glucose uptake of the liver cells, this dose given intraportally might be followed by an alteration in the net amount of glucose leaving the liver, and also in its release of unlabeled glucose. This, however, was not the case, as the values remained approximately constant for the whole infusion period, although the hepatic cells presumably were exposed to concentrations of insulin that would have stimulated the glucose uptake of peripheral tissues.

Similar results were obtained with intraportal infusion of 0.04, 0.08 and 0.12 I.U./kg./hour. The decrease in the arterial blood sugar indicates only an increased uptake of glucose in the peripheral tissues, as the output from the liver remained almost constant in this period. Thus, the results obtained after intraportal infusion of relatively small doses of glucagon-free insulin into cyclopropane-anesthetized normal dogs indicate that the glucose output from the liver is not even partly inhibited by the influence of exogenous insulin.

Discussion

The temporary stop in the decrease of the plasma glucose specific activity following tolbutamide injections into normal dogs and, more so, the longer lasting plateau observed in the pancreatectomized dogs are thought to be due to an inhibition of the output of unlabeled glucose from the liver. A similar inhibitory action of the sulfonylureas upon the hepatic release of glucose has been observed by Anderson *et al.*,[7] Recant and Fischer,[8] and Ashmore *et al.*[9] Simultaneously, a stimulatory effect upon the glucose uptake of the peripheral tissues could not be demonstrated, which is in [illegible] and Yalow[10] and of Ashmore *et al.*[9] [illegible] er as a single intravenous injection [illegible] plasma glucose concentration was never associated with any decrease in the hepatic glucose output. Thus the stimulatory effect of insulin upon the glucose uptake in the periphery was responsible for the fall in the blood sugar. This was also evident from the increased rate at which the plasma glucose specific activity decreased after the insulin was administered, and confirms previous observations with C^{14} glucose.[9, 10]

Conclusions

As the hypoglycemic effect of the sulfonylureas is not associated with any such stimulation of the glucose uptake, it must be concluded that their action is not likely to be exerted via an increase in the secretion of insulin from the pancreas or in the amount of insulin available in the body, but must be due

to its inhibitory effect upon the glucose output from the liver However as Dulin and Johnson[11] and Sobel *et al* [12] have been able to demonstrate a hypoglycemic effect of sulfonylureas in the hepatectomized rat or dog the action upon the hepatic glucose output may not be the only mode of action of these compounds but it presumably plays a major role in the intact animal which is not able to mobilize more liver glycogen to counteract the hypoglycemia produced by the sulfonylureas

Summary

The influence of tolbutamide and insulin upon the hepatic glucose output and the glucose uptake in the peripheral tissues was studied in normal and pancreatectomized dogs and diabetic patients by applying the technique of single injection of C^{14} glucose

Tolbutamide produced a definite cessation in the constant dilution of the plasma glucose specific activity normally caused by the release of unlabeled glucose from the liver to replace the blood sugar taken up by the periphery as demonstrated by a plateau in the specific activity curve which was most pronounced in the pancreatectomized dogs and the diabetic patients As no increase in the peripheral utilization of glucose was demonstrated in these preparations it was concluded that the hypoglycemic effect of tolbutamide is caused by an inhibition of the release of glucose from the liver

By combining hepatic venous catheterization with the C^{14} gluco e technique a new method of calculating the hepatic glucose output was worked out this was not affected by changes in the size of the glucose pool or the [illegible]

1 U /kg /hour In no instance was this followed by a decrease in the hepatic glucose output the hypoglycemia was solely produced by a stimulation of the peripheral glucose utilization which is not observed after injections of tolbutamide

It was therefore concluded that it is very unlikely that the sulfonylureas exert their hypoglycemic effect via an increased release of insulin from the pancreas

Acknowledgments

The authors express their indebtedness to K Hallas Møller for many valuable suggestions and stimulat ng criticism and to A Thordahl Christensen The Royal Veterinary and Agricultural College Copenhagen for placing at their disposal the X ray facilities of his department The technical assistance of Kirsten Harder and of Lise Wollesen is also gratefully acknowledged

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Discussion of the Paper

SIDNEY WEINHOUSE (*The Institute for Cancer Research, Philadelphia, Pa*) I appreciate this opportunity of describing some experiments published in the September October 1958 issue of *Diabetes* These were carried out by my colleagues Reichard Jacobs and Friedmann Like the studies already reported by James Craig and Leonard Madison they suggest an important role for insulin, as well as for the synthetic hypoglycemic agents, in suppressing hepatic glucose output

FIGURE 1 shows results of an experiment similar to those that have just been described by Schambye, indicating that in the normal human hepatic glucose output is suppressed by the intravenous injection of insulin in a dosage of 0 1 U /kg With the administration of insulin there is an immedi ate drop in the blood sugar which coincides exactly in time with the

by a resumption in drop of specific activity ...
begins to level off Approximate inflow and outflow rates can be calculated from the data presented this is shown in the inset of the figure The solid line represents the inflow rates the dotted line represents the rate of disappearance presumably due to peripheral utilization When the insulin is administered in one dose there is a profound lowering or disappearance of glucose inflow simultaneous with a large increase in glucose disappearance rate

FIGURE 2 shows a similar experiment in which tolbutamide was administered Experiments were not carried out with chlorpropamide, unfortunately, but we hope to do this Here again it can be seen that, coinciding with the hypoglycemic action of the drug, there is a "plateauing" of specific activity of the blood glucose for a period of about 20 to 30 min , again indi

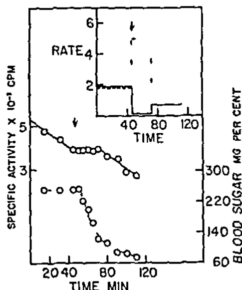


FIGURE 1 Blood sugar changes in a diabetic patient weighing 60 kg and given 6 U of insulin (indicated by arrow). Broken line represents blood glucose level; solid line, specific activity. Inset shows approximate values with solid line representing entry, broken line removal.

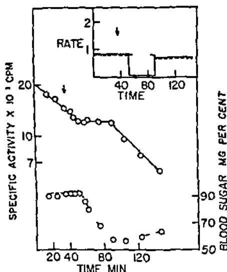


FIGURE 2 Blood sugar changes in a nondiabetic patient weighing 50 kg and given 1.2 gm tolbutamide intravenously (indicated by arrow). Broken line represents blood glucose level; solid line, specific activity. Inset shows approximate rates with solid line representing entry, broken line removal.

cating a suppression of glucose output However, when the inflow and outflow rates are calculated, we find a significant difference from the experiments with insulin

Although tolbutamide lowers the inflow of blood glucose, it does not have the stimulating effect on utilization observed when insulin was given This apparent lack of stimulation of peripheral utilization represents a profound difference in the action of the two hypoglycemic agents

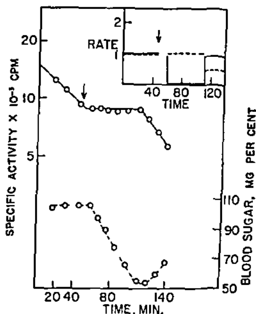


FIGURE 3 Effect on the time course of blood glucose concentration of 10 U of insulin given subcutaneously to a nondiabetic patient weighing 66 kg Arrow indicates time of administration of insulin Broken line represents blood glucose level, solid line specific activity Inset shows approximate rates with solid line representing entry, broken line, removal

It also occurred to us, as it did to other investigators, that endogenous insulin secreted slowly into the portal vein might have an effect different from that of intravenously injected insulin To simulate the slow release of insulin that, presumably, takes place when tolbutamide is injected, we did some experiments in which we injected the same amount of insulin subcutaneously, so that there would be slow release into the blood stream

inflow without a simultaneous increase in the utilization of glucose

We interpret these results as indicating that the slow introduction of small doses of insulin into the blood stream causes a depression of the hepatic glucose output, but has little or no effect on the peripheral utilization of

glucose We have also carried out experiments using slow intravenous infusion, and we have obtained identical results

As others have indicated, these results do not exclude the possibility that tolbutamide or chlorpropamide have effects other than the stimulation of insulin release, but they do reconcile some of the previous results that were interpreted as indicating a different effect More significantly, these results suggest to us that we should reconsider and re evaluate the possibility of hepatic glucose output regulation in the problem of diabetes

COMPARISON OF SOME METABOLIC EFFECTS OF INSULIN, CHLORPROPAMIDE, AND OTHER HYPOGLYCEMIC SUBSTANCES*

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Convincing experimental evidence suggests that carbutamide and tolbutamide stimulate the release of insulin from the pancreas [1, 16] However, this cannot be accepted as the sole explanation for the hypoglycemia because these drugs do not share other metabolic effects of exogenous insulin [8, 21] Two hypotheses may explain these differences in mode of action: (1) insulin secreted into the portal system as a result of normal pancreatic function or pancreatic stimulation acts differently from insulin injected peripherally, or (2) the drugs act not only on the pancreas but on the liver[13, 27] or other tissues also Experiments designed to study these hypotheses have given contradictory results Two groups of workers compared the effects of insulin and tolbutamide on the rate of disappearance from the plasma of injected C^{14} glucose The first group[23] concluded that tolbutamide causes hypo

insulin injected into the femoral vein The other group of workers [4] confirmed the observation concerning tolbutamide and insulin injected intrave-
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heral vein, thus
confirming the observation that the metabolic effects of insulin may vary, depending upon the route of administration Unfortunately, these conclusions are based on the measurement of arteriovenous (A-V) differences without measurements of blood flow Experiments performed in this labora-
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made and tolbutamide may act on the liver but only if small (permissive) quantities of endogenous or exogenous insulin are provided [26, 27] Essentially the same conclusion was reached recently by other investigators [25] This paper reports similar observations on the action of chlorpropamide

Methods

rapid induction and the long lasting uniformly deep anesthesia with complete muscle relaxation that is essential to prevent fluctuations in blood pyruvate and lactate unrelated to the experiments Chlorpropamide* was injected rapidly into the femoral vein in doses of 5 or 50 mg /kg or into the pancreatic artery in doses of 5 mg /ml /kg at the rate of 1 ml /min Control injections of saline into the femoral vein and of saline or glucose (0 5 gm in 20 cc) into the pancreatic artery had been performed previously [27, 28] Blood samples were taken from an exposed femoral vein for duplicate analyses Glucose was determined according to Nelson [30] pyruvate according to Friedemann and Haugen [1] lactate according to Barker and Summerson [3] and potassium with the Coleman flame photometer The carotid pressure of the dogs was used to check their general condition which remained satisfactory in all but two animals these were discarded The statistical significance of the results was calculated according to Fisher [32]

Results and Discussion

The results are given in the form of curves representing average changes from preinjection values FIGURE 1 shows that the injections of saline or chlorpropamide in doses of 5 mg /kg into the pancreatic artery or into the femoral vein had no significant effect on peripheral blood glucose concentration In contrast the injection of glucose caused a significant ($p < 0.01$) hypoglycemia The results suggest that contrary to the results obtained under comparable conditions with carbutamide and tolbutamide chlorpropamide does not stimulate insulin release FIGURE 2 shows that the injection of chlorpropamide in doses of 50 mg /kg into the femoral vein causes a prompt and significant decrease in the concentrations of blood glucose ($p < 0.01$) and ...
($p < 0.05$) d
cant effect or

not designed to determine whether the hypoglycemic action of chlorpropamide like that of carbutamide and tolbutamide requires the presence of small (permissive) quantities of insulin

* Donated by Chas Pfizer & Co Inc Brooklyn N Y

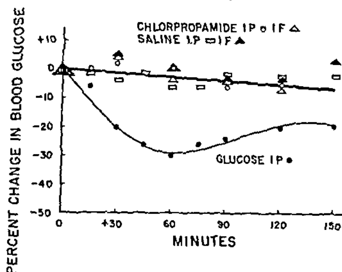


FIGURE 1 Effect of intrapancreatic (I P) or intrafemoral (I F) inject ons of chlor propamide or sal ne (stra ght line) and of intrapancreatic inject ons of glucose (curve) on blood glucose

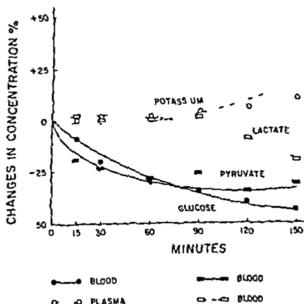


FIGURE 2 Effect of intrafemoral inject ons of chlorpropamide on blood glucose pyruvate lactate and plasma potass um

Summary and Conclusions

Chlorpropamide was injected into the pancreatic artery or into the femoral

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tions of blood glucose pyruvate and lactate The results are consistent with the hypothesis that the hypoglycemic effect of the drug is due at least in part, to a decreased liver glucose production and offer no evidence for a pancreatropic action of chlorpropamide

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EFFECT OF CHLORPROPROPAMIDE ON INTERMEDIARY METABOLISM IN HUMAN SUBJECTS*

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Development of the oral hypoglycemic agents has stimulated widespread investigation in an attempt to clarify their mode of action As a consequence, the metabolic activities of insulin have been investigated more intensively At the present time there are two general concepts about the action of the sulfonylureas One concept is that the principal action of tolbutamide and carbutamide is to stimulate insulin release from the pancreas[1 2] the other is that these agents produce hypoglycemia by decreasing hepatic glucose output without increasing peripheral utilization[3 7] Almost all metabolic studies reported in human and in experimental animals have been acute experiments, that is, the metabolic effects have been studied over a period of a few hours after the administration of the initial dose of the drug It was felt that it would be worthwhile to investigate the metabolic effects of chronic administration of chlorpropamide in diabetic patients who were being maintained on it

Methods and Materials

Diabetic patients were selected from the general Diabetic Clinic at Jefferson Davis Hospital, a city county charity institution Patients known to be free of metabolic disease and members of the professional staff served as normals Where possible, studies were performed before as well as after chlorpropamide therapy was instituted Each patient had received chlorpropamide at least one week before the "on treatment' study was performed After an over night fast the patients came to the metabolic laboratory and remained at rest in the laboratory until completion of the test They took insulin and/or chlorpropamide in the dose of 500 to 1500 mg /day in their usual manner Blood samples were obtained while the patient was fasting and at 1, 2 and 4 hours after administration It is known that the composition of the diet will influence the blood levels of certain metabolites It would be desirable to have all patients on a standard diet, but this is not possible because of the very limited resources available to this group of patients Experience with these subjects has shown that the majority consume a rather constant diet that is low in protein and high in carbohydrate fat, and calories,[8] for this reason no dietary instructions were issued and the patients were asked to remain on their usual diets

In the majority of published studies glucose has been used as the test meal In order to determine whether any other abnormalities would appear when a

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more normal dietary mixture was used the bulk of the data herein reported was obtained using a test breakfast containing 20 gm protein 65 gm carbohydrate and 20 gm fat Additional studies utilizing glucose have also been performed for comparison Analyses were performed for blood sugar,[9] inorganic phosphate [10] pyruvate [11] alpha ketoglutaric acid [11] lactate [12] total ketones [13] and citrate [14]

Results

The blood sugar lowering effect of chlorpropamide is seen in FIGURE 1 It should be noted that after the test meal the normals did not develop any elevation in blood sugar which is probably due to delayed absorption In contrast the diabetic developed significant elevations both before and after

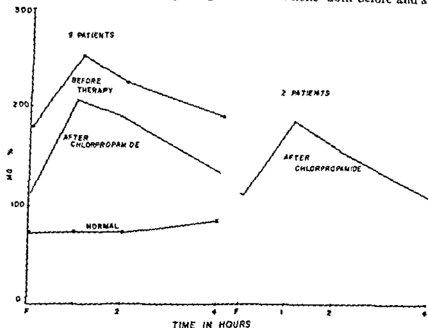


FIGURE 1 Blood sugar after test meal

therapy However the after therapy curve is significantly lower than the before therapy curve although its shape is not altered greatly In general these results are in agreement with those reported by other investigators utilizing tolbutamide and carbutamide in acute experiments [3,7]

The studies on serum inorganic phosphate appear in FIGURE 2 In spite of the flat glucose tolerance curve the normals showed typical fall in phosphate following the meal and the values returned to normal by the fourth hour In the insulin treated diabetics this response is still seen, but is less marked In the chlorpropamide treated diabetics there is no change in serum phosphate level following the test meals Statistical analysis indicates that there is no difference in the results obtained before and after therapy These find

ings are in agreement with other reports[3 5 7] on the effect of tolbutamide and carbutamide after acute administration

Pyruvate blood levels (FIGURE 3) normally rise after a meal with fall back to normal at 2 to 4 hours [15 16] The same is true of the insulin treated diabetic The untreated diabetic also has essentially the same response The chlorpropamide treated diabetic however has an elevated fasting level of pyruvate that fell after ingestion of the meal This elevation is of borderline significance and appears in all of the experiments This fasting elevation of pyruvate in chronically treated patients has not been reported previously The data of Hennes *et al* [14] are not comparable, as these authors subjects were

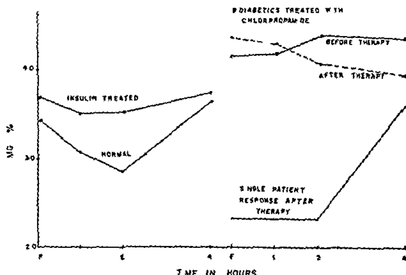


FIGURE 2 Inorganic phosphate

not receiving sulfonylureas on a chronic basis The elevation in pyruvate if substantiated would indicate either increased production probably from gluconeogenesis or decreased utilization As there has been no evidence that sulfonylureas increase gluconeogenesis it is probable that the elevated fasting pyruvate level is due to decreased utilization of pyruvate

Under the conditions of these studies pyruvate lactate citrate and alpha-ketoglutarate will vary considerably, and alterations would require rather considerable magnitude to be significant The results on lactate appear in TABLE 1 and show that there is no significant alteration although there is considerable variability The relatively high fasting values are probably due to recent activity of the patient in coming to the hospital rather than any drug induced effect [17] In TABLE 2 it is seen that after the test meal the citric acid level rises at the first hour and then falls to normal Other investigators have shown that the response of serum citrate in normal persons

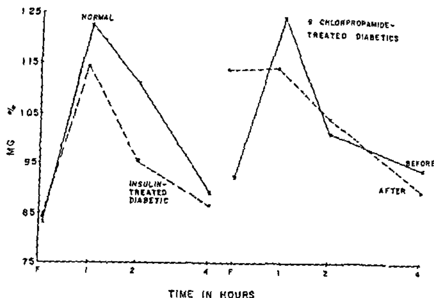


FIGURE 3 Pyruvate

after glucose feeding is a fall with a return to normal in 4 to 6 hours [11, 15] However, in diabetics there is often a rise after the meal Glucose was fed to 6 normal individuals, and the serum citric acid levels fell as described in the literature [18] It appears that the rise in citrate in normals after the test meal *is related to the diet* *In diabetics, before treatment, the postprandial rise* in citrate is rather large and prolonged, and this is not altered after treatment with either insulin or chlorpropamide The studies on alpha ketoglutarate (TABLE 3) reveal no striking changes The apparent elevated fasting levels after chlorpropamide therapy in the group that was studied before and after therapy would appear to approach significance, but the results obtained in the group studied only after chlorpropamide therapy do not support this

TABLE 1
LACTATE
(Time in hours)

	F	1	2	4	No pts
	Mean ± S E mg %				
Normal	11 1 ± 1 2	9 3 ± 0 62	9 3 ± 0 68	9 5 ± 1 13	6
Insulin	*11 8 ± 1 3*	12 9 ± 1 7	11 3 ± 1 5	11 8 ± 1 9	8
Chlorpropamide					
Before	10 9 ± 1 1	12 04 ± 1 3	12 5 ± 1 9	10 6 ± 1 4	12
After	9 9 ± 0 58	10 4 ± 0 66	10 8 ± 0 91	10 0 ± 0 75	15

TABLE 2
CITRIC ACID
(Time in hours)

	F	1	2	4	No pts
	Mean ± S E mg %				
Normal	2 1 ± 0 28	2 2 ± 0 16	2 01 ± 0 24	1 82 ± 0 13	6
Insulin	2 1 ± 0 13	2 8 ± 0 19	2 9 ± 0 22	2 6 ± 0 23	8
Chlorpropamide					
Before	2 6 ± 0 09	3 0 ± 0 25	3 2 ± 0 15	2 6 ± 0 10	9
After	2 3 ± 0 12	2 9 ± 0 24	2 8 ± 0 14	2 5 ± 0 23	9

TABLE 3
ALPHA KETOGLUTARIC ACID
(Time in hours)

	F	1	2	4	No pts
	Mean ± S E mg %				
Normal	0 45 + 0 09	0 52 ± 0 18	0 35 + 0 15	0 36 ± 0 14	6
Insulin	0 47 ± 0 10	0 69 ± 0 12			13
Chlorpropamide					
Before	0 41 ± 0 06	0 51 ± 0 07	0 40 ± 0 05	0 36 ± 0 07	9
After	0 54 ± 0 13	0 48 ± 0 05	0 46 ± 0 05	0 40 ± 0 05	9
After unpaired	0 44 + 0 07	0 45 ± 0 03	0 35 ± 0 06	0 33 ± 0 05	12

TABLE 4
KETONES
(Time in hours)

	F	1	2	4	No pts
	Mean ± S E mg %				
Normal	1 95 ± 0 19	1 75 ± 0 48	2 17 ± 0 5	2 9 ± 0 93	6
Insulin	3 15 ± 0 56	2 24 ± 0 49	2 75 ± 0 66	2 8 ± 0 59	8
Chlorpropamide					
Before	4 62 ± 1 04	3 04 ± 0 61	2 83 ± 0 50	3 13 ± 0 68	12
After	2 95 ± 0 77	2 20 ± 0 31	2 08 ± 0 26	2 31 ± 0 30	12

The blood ketone levels appear in TABLE 4 It may be seen that there is *increased ketone production in the untreated diabetic* This is returned to a more normal level by the administration of insulin or chlorpropamide The insulin treated diabetics herein reported were in only fair control, and the chlorpropamide treated patients had been on this drug for approximately 1 to 2 weeks With better control of the disturbed carbohydrate metabolic defect in diabetics the production of ketones is further diminished for example a study of 6 patients who were in excellent control after several months of chlorpropamide therapy showed a mean fasting blood ketone level of 0 89 ± 0 18 (S E) *This does indicate that the chlorpropamide has the* ability to increase the utilization of carbohydrate to the point of reducing ketone production to low normal

Conclusions

The study of human intermediary metabolism in normal and diabetic patients before and after administration of insulin and chlorpropamide indicate that there are some differences between the metabolic action of insulin and chlorpropamide The configuration of the glucose curve and the failure of the phosphate response indicate that chlorpropamide on chronic administration has essentially the same action as that of tolbutamide and carbutamide *and that this action differs from that of insulin In addition* there is an elevation of fasting pyruvate level after the administration of chlorpropamide The chronic administration of chlorpropamide effectively suppresses overproduction of ketones *No significant alterations were seen in citrate* lactate and alpha ketoglutaric blood levels however small differences would not appear under the conditions of these experiments

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METABOLIC EFFECTS OF CONTINUED ADMINISTRATION OF SULFONYLUREA DERIVATIVES IN SELECTED DIABETIC SUBJECTS INTERRELATIONS WITH GLUCAGON*

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Although the mode of action of sulfonylureas is not completely understood, certain well established facts have emerged from the accumulated clinical and experimental data For example several laboratories have demonstrated that the hypoglycemic action of sulfonylurea compounds is associated with a reduced output of glucose by the liver [1-5] The mechanisms by which the reduced glucose output is accomplished have not been clearly established despite an intensive study of the problem However, a decreased conversion of galactose or fructose to glucose by the liver has been observed during the period of hypoglycemia induced by administration of sulfonyl ureas [6, 7] and this has suggested the possibility that selective interference with hepatic gluconeogenesis might contribute significantly to the hypoglycemia Little if any information is available on the effect of sulfonyl ureas on conversion of amino acids to glucose in the liver

In the present studies an attempt has been made to determine whether the sulfonylureas might inhibit hepatic gluconeogenesis from protein and amino acids as well as hepatic glycogenolysis To this end the effect of sulfonyl ureas on the metabolic responses to repeated daily intramuscular injections of glucagon has been studied The ability of glucagon to increase the daily urinary nitrogen excretion as well as to raise the daily blood and urinary sugar levels with and without the simultaneous administration of carbutamide has been studied in two diabetic patients† who exhibited a striking hypoglycemic response to carbutamide The effects of glucagon and carbutamide on the daily fasting levels of serum inorganic phosphorus on the daily urinary excretion of reducing corticosteroids and on the daily balances of phosphorus potassium and sodium have also been investigated

The rationale for the glucagon carbutamide experiments was provided by earlier work in this laboratory Metabolic studies with fed diabetic patients[8, 9] and with fasting experimental rats[10] have shown that increased protein catabolism seems to be an integral part of the hyperglycemic glycogenolytic response to fairly large doses of glucagon Furthermore, studies of the rat have suggested that the increased protein catabolism induced by

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† The results of such a study in one of the patients have been published in part in a preliminary report [9]

glucagon is probably due to an enhanced hepatic gluconeogenesis from amino acids

The second part of this paper will deal with the question of a possible hypoglycemic action of sulfonylureas in the severe, unstable diabetic receiving suboptimal doses of exogenous insulin There is well-documented evidence showing that, in long-term studies, carbutamide and tolbutamide can lower the levels of glucose in blood and urine in depancreatized dogs receiving suboptimal doses of exogenous insulin,[11 14] but not in severe, unstable diabetes[15 17] in humans similarly treated with suboptimal doses of exogenous insulin With the introduction of more potent hypoglycemic agents such as chlorpropamide, it was considered worthwhile to reinvestigate this problem The effects of tolbutamide and chlorpropamide on carbohydrate and protein metabolism have been determined for two patients with severe, unstable, long standing diabetes, who were presumed to be critically lacking in endogenous insulin

Methods

Subjects

Four patients with diabetes mellitus served as subjects One of the patients (R E) had the stable type of diabetes, the remaining three had the unstable type The illness was of very recent onset in one of these unstable cases (A F), in the other two cases (E R and E D) it was of many years' duration The subjects were classified as unstable or stable according to a criterion described in a previous paper[18] Throughout the period of study, which in each case extended several months, the patients lived quietly in the metabolism ward of Strong Memorial Hospital under routine but rigidly controlled conditions Physical activity was usually restricted to being up and about in the ward as the patients wished

Diets

Each patient received a weighed diet constant in chemical composition, the identical menu being served every day The methods and rationale for calculating protein, fat, carbohydrate, total calories, and distribution of carbohydrate among the meals have been reported elsewhere[19]

Insulin

Insulin was administered regularly to each patient as a single daily dose of either NPH or lente insulin every morning before breakfast Initially, the patients were placed on approximately the same dose of insulin that they had been receiving before hospitalization Subsequently, the dose was adjusted to achieve optimal regulation of blood and urinary sugar levels In order to achieve more satisfactory regulation of the levels of sugar in blood and urine patients A F, E R, and E D received also a small dose of crystalline insulin each morning before breakfast Suboptimal regulation of blood and urinary sugar levels was achieved by appropriate reduction of the total daily dose of insulin to the point where the levels of blood and urinary sugar became

significantly higher than those obtained with optimal doses of insulin, b without the development of ketosis

Analytical Procedures

All patients were weighed daily every morning before breakfast, and fastin blood sugar and 24 hour urinary excretion of sugar, nitrogen, phosphoru potassium, and sodium were determined daily for each A sample daily men for each patient was analyzed for nitrogen, phosphorus, potassium, an sodium In each case the average daily fecal excretion of nitrogen, phos phorus, potassium, and sodium was calculated from a pooled 5-day collectio of feces Carmine was used to mark the beginning and end of the period o collection Daily balances for nitrogen, phosphorus, sodium, and potassiun were obtained by subtracting the observed daily 24 hour urinary values anc calculated average daily fecal values from measured amounts of nitrogen phosphorus, sodium, and potassium in a sample diet Fasting plasma, inor ganic phosphorus, and 24 hour urinary excretion of reducing corticosteroids were determined daily in patients R E and A F In patient R E the fasting blood levels of carbutamide and capillary blood sugar were also meas ured daily

Capillary samples of blood were drawn from the finger tip, usually in dupli cate within 1 or 2 min of the venipuncture using pipettes calibrated to con tain 0 2 ml The purpose of this procedure was to estimate the concentration of glucose in arterial blood (capillary blood was assumed to equal arterial blood in glucose concentration) The fasting levels of total plasma amino acids and chlorpropamide were determined daily in patient E D

Chemical Methods

Sugar in blood and urine was determined by the Shaffer Hartman copper iodometric method,[20] using Somogyi s new copper reagent [21] Urines were analyzed for total nitrogen in duplicate by a modified micro Kjeldahl method Diets and stools were analyzed for total nitrogen in triplicate by the macro-Kjeldahl method using the wet" technique [22] Sodium and potassium in diets, stools, and urines were determined by means of a Baird flame photome ter Inorganic phosphorus in plasma urine diet, and stool was measured by the method of Fiske and SubbaRow [23] Total reducing corticosteroids in urine were determined as follows the urine was hydrolyzed for 48 hours at 47° C with β-glucuronidase after adjustment to pH 4 5 and buffering After the steroids had been extracted in chloroform a ' neutral extract of the latter[24] was prepared Finally, the corticosteroids were estimated colorimet rically by a modification of the Mader and Buck method[25] using blue tetra-zolium (BT) [26] Plasma amino acid nitrogen was measured by the method of Frame *et al* [26] Serum carbutamide levels were measured by a modification of the method of Marshall and Bratton [27] and serum chlorpropamide levels were determined by D Iezzoni of Chas Pfizer & Co , Inc , Brooklyn, N Y.

Glucagon

Patients R E and A F were allowed to reach nitrogen equilibrium before the control data were obtained After suitable periods (of several days'

duration) each patient received either 1 or 2 mg of an aqueous solution of glucagon intramuscularly every 8 hours 24 hours per day for 6 days One such course of glucagon was administered during the period of optimal regulation of blood and urinary sugar levels with exogenous insulin as described above and one course was given during optimal regulation of sugar levels with carbutamide The glucagon used in these studies was an approximately 50 per cent pure amorphous preparation * It contained between 0 005 and 0 05 U of insulin per milliliter of solution and bore the Lot No 208 158B 214A

Arylsulfonylureas

All of the sulfonylureas used in these studies were given by mouth daily in divided doses Patients K E and A F received 2 0 gm and 2 5 gm of carbutamide * respectively per day Patient E R received 1 5 to 2 5 gm of tolbutamide† per day, and patient E D was given 2 separate courses of chlorpropamide ‡ the first at 1 5 gm per day and the second at 2 0 gm daily At least 2 or 3 days were allowed to elapse after beginning or stopping treatment with the drug before the metabolic data were obtained

Results

Effects of Carbutamide on the Responses of Various Metabolic Indices to Glucagon

The pertinent metabolic data obtained in patients R E and A F are shown in FIGURES 1 and 2 and in TABLES 1 and 2 respectively All data are expressed as mean daily values ± 2 standard errors about the mean with the exception of the $\frac{A I}{I}$ ratios in FIGURE 1 The latter values are expressed as the mean daily values ± 1 standard error about the mean This was done to avoid negative values about the mean which would have occurred for some periods because of the marked variability in the daily observed values and because of the small size of the sample

Blood and urinary sugar levels Before comparing the responses of the blood and urinary sugar levels to glucagon when the patients were or were not receiving carbutamide it may be well to point out that administration of carbutamide caused a striking fall in the levels of fasting blood sugar and in the excretion of glucose This is evident from a comparison of the blood and urinary sugar levels in periods 1 and 5 with those of periods 6 and 8 in FIGURE 1 and of the levels in period 4 with those of periods 5 7 and 8 in FIGURE 2 In patient R F the regulation of both the fasting blood sugar and the amount of sugar excreted in the urine was equal if not superior to that achieved by the administration of 36 U of insulin daily and in patient A F the regulation by carbutamide was as good as or better than the administration of 100 U of insulin daily without carbutamide In both cases the daily ingestion of 2 0 to 2 5 gm of carbutamide resulted ultimately in fasting blood sugar levels close to or within the normal range and the excretion of less than

* Supplied by Eli Lilly and Company Indianapolis Ind
† Supplied by The Upjohn Company Kalamazoo Mich
‡ Supplied by Chas Pfizer & Co Inc Brooklyn N Y

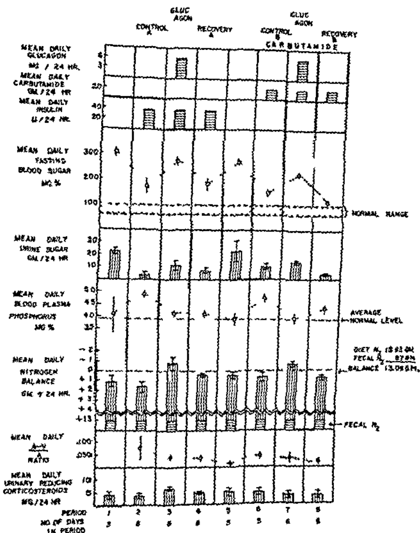


Figure 1. Effect of continued administration of carbutamide on the various metabolic responses to intramuscular injections of glucagon every 8 hours for 6-day periods. All data are expressed as mean daily values ±2 S.E. about the mean except for the $\frac{A-V}{A}$ ratio which is expressed as the mean daily fasting ratio ±1 S.E. about the mean. For further explanation see text. Patient R. F. female 64 years old, stable diabetes long duration.

2.0 gm. of glucose in the urine every 24 hours. The mean daily level of blood carbutamide was 7.7 mg. per cent in patient R. E. and 7.14 mg. per cent in patient A. F. It should be noted that there was greater consistency in fasting blood sugar levels from day to day when the patients were receiving carbutamide than when they were receiving insulin only, as shown by the

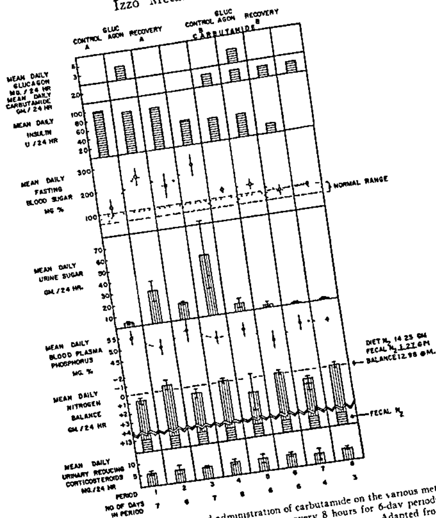


FIGURE 2 Effect of continued administration of carbutamide on the various metabolic responses to intramuscular injections of glucagon every 8 hours for 6-day periods All data are expressed as mean daily values ±2 S E about the mean Adapted from Izzo and Roncone[4] (by permission of *Diabetes The Journal of The American Diabetes Association*) Patient A I female 19 years old unstable diabetes short duration

fact that a smaller standard error was obtained with carbutamide than with insulin Moreover the need for insulin in both patients is evident from the levels of hyperglycemia and the glycosuria that resulted when insulin was omitted (periods 1 and 5, FIGURE 1) or reduced to suboptimal levels (period 4, FIGURE 2)

Intramuscular injections of glucagon administered to both patients during

Table 1
Phosphorus Sodium and Potassium Balance Data of Patient R F Female 64 Years Old Stable Diabetes of Long Duration*

	P r d							
	1	2	3	4	5	6	7	8
No of days	4	6	6	5	5	(	6	5
Carbutamide gm /24 hours								
Glucagon mg /24 hours		6				2 0	2 0	2 0
Insulin U /24 hours		16	36	36			0	
Urine inorganic phosphorus gm /24 hours	6283 ± 058	568 + 110	680 ± 066	597 ± 124	396 ± 134	510 ± 216	729 ± 028	652 ± 058
Urine sodium mEq /24 hours	63 71 ± 12 0	40 2 ± 5 0	54 8 ± 13 4	43 2 ± 6 8	61 0 ± 5 4	63 4 + 9 2	61 8 ± 5 6	51 4 ± 8 0
Urine potassium mEq /24 hours	74 41 ± 7 2	77 6 ± 8 6	87 2 ± 13 8	84 2 ± 3 6	87 4 ± 7 4	80 5 ± 7 0	79 7 ± 1 4	87 5 ± 2 4

* P r further deta ls see text and FIGURE 1 All urinary alues are expre sed as the mean ± 2 S E ab ut the mean
† T obta n the da ly balance subtract the urinary value from the a erage da ly intake Ur nary alues less than the average daily ntake nd cate pos t ve bal ance and ur nary values greater than the average daily ntake indicate negative balance

	Phosphorus gm	Potass um mEq	Sod um mEq
Sample da ly d et	1 606	92 04	55 15
Average da ly fecal excret on	− 686	− 4 16	− 1 93
A erage da ly ntake	920	87 98	56 22

TABLE 2

PHOSPHORUS SOD UM AND POTASSIU M BALANCE DATA OF PATIENT A 1 FEMALE 19 YEARS OLD UNSTABLE DIABETES OF SHORT DURATION*

	P riod							
	1	2	3	4	5	6	7	8
No f days	7	6	7	7	3	6	4	2
Ca l utam de gm /24 hours					2 5	2 5	2 5	2 5
Glucagon mg /24 hours		3 6				3		
Insul n U /24 hours	100	90	90	54	50	50	35	—
Ur ne norgan c phosphorus gm /24 hours	950† ± 136	986 ± 104	890 ± 174	1 061 ± 132	945 ± 114	1 008 ± 074	912 ± 176	921 ± 138
Ur ne sod um mEq /24 hours	64 5† ± 21 4	63 3 ± 28 0	62 0 ± 29 0	65 5 ± 27 0	64 6 ± 5 8	64 2 + 16 8	63 4 ± 19 8	66 9 ± 38 6
Urine potass um mEq /24 hours	62 7† ± 3 0	59 1 ± 5 2	61 0 + 5 6	68 8 ± 8 4	63 0 ± 4 2	75 3 ± 6 2	69 9 ± 18 8	9 7 ± 1 2

* For further deta ls see text and F GURE 2 All urinary alues are expressed as the mean ± 2 S D about the mean

† To obta n the da ly ba ance subtract the urinary value from the average da ly ntake urinary values less than the average da ly ntake and cate pos t ve bal an e and urinary values greater than the ave age da ly ntake nd cate negat e balance

	Phosphorus gm	Potass um mEq	Sod um mEq
Sample da ly d et	1 377	82 38	0 46
Average da ly fecal excretion	– 540	18 31	6 39
Average da ly intake	837	64 07	64 07

the time of optimal regulation with exogenous insulin resulted in sharp increases in fasting blood sugar levels and 24 hour excretion of sugar (periods 2 and 3, FIGURE 1, and periods 1 and 2, FIGURE 2) The mean increase in daily fasting blood sugar above control levels was 101 mg per cent in patient R E and 119 mg per cent in patient A F The mean increases in daily urinary sugar levels were 7 31 gm and 24 47 gm, respectively With discontinuation of glucagon the levels of blood and urinary sugar promptly moved toward the control levels (period 4 FIGURE 1 and period 3, FIGURE 2) Intramuscular administration of glucagon to patient R E during the time she was receiving carbutamide resulted in a mean increase in fasting blood sugar level, which was only slightly less than that caused by similar injections of glucagon during the time of optimal insulin regulation (periods 7 and 8 FIGURE 1) The mean rise in 24 hour excretion of sugar was 3 53 gm It should be noted that the rise in the levels of fasting blood sugar induced by glucagon when patient R E was receiving either insulin or carbutamide was not associated with any increase in peripheral glucose utilization, as measured by the $\frac{I\ I}{A}$ ratio In sharp contrast to the results obtained in patient R E, the administration of glucagon to patient A F at a time when she was receiving carbutamide with reduced doses of insulin produced no rise in levels of blood or urinary sugar (periods 5, 6, and 7, FIGURE 2)

glucose in the blood and urine glucagon also caused a significant rise in the excretion of nitrogen in the urine, with a reversal of nitrogen balance from positive to slightly negative (period 3 FIGURE 1 and period 2, FIGURE 2) The mean daily increase in the excretion of nitrogen during the period of glucagon administration was 2 39 gm higher than the control levels in patient 1 and 1 40 gm higher than the control levels in patient 2 With discontinuance of glucagon the excretion of nitrogen promptly decreased and the nitrogen balances again became positive (period 4, FIGURE 1 and period 3, FIGURE 2) It should be noted that patient R E was in positive nitrogen balance during periods 1 and 5, FIGURE 1 when insulin was omitted, even though the levels of hyperglycemia and glycosuria were similar to or even higher than the levels produced by glucagon and insulin Glucagon also caused a greater excretion of nitrogen in patient A F than did reduction of insulin to suboptimal levels although the hyperglycemia and glycosuria were greater during the period of suboptimal insulin administration than during the period of administration of the optimal amount of insulin together with glucagon

The administration of glucagon in the presence of carbutamide also resulted in a significant increase in the excretion of nitrogen in the urine in patients R E and A F The rises were roughly of the same order of magnitude as the rise in nitrogen excretion that occurred when the patients were given glucagon while receiving optimal doses of insulin Nitrogen balances

reverted from positive to slightly negative in patient R E, and to equilibrium in the case of patient A F (periods 6 and 7, FIGURE 1, and periods 5 and 6 FIGURE 2) *The increase in the mean daily nitrogen excretion induced* by administration of glucagon was 1 29 gm in patient R E and 1 67 gm in patient A F As in the glucagon insulin studies nitrogen excretion promptly decreased and the nitrogen balances again became positive when the injections of glucagon were stopped (period 8 FIGURE 1 and period 7 FIGURE 2) It is of interest to note that carbutamide did not diminish the nitrogen response to glucagon in patient A F even though it virtually abolished the hyperglycemic glycosuric response to glucagon Nitrogen retention was greater in patient A F when she was receiving carbutamide and reduced doses of insulin than when carbutamide alone was given although the regulation of blood and urinary sugar levels in both periods 7 and 8 FIGURE 2, was similar However because periods 7 and 8 were of short *duration*, little significance can be attached to these differences in nitrogen excretion

Plasma inorganic phosphorus Before discussing the effect of carbutamide on the response of the level of plasma inorganic phosphorus to the administration of glucagon it is pertinent to point out the occurrence of an inverse relationship between the level of fasting blood sugar and the level of fasting plasma inorganic phosphorus in both patients R E and A I This is shown in patient R E by the significant rise in mean daily level of plasma inorganic phosphorus from 4 11 mg per cent to 4 88 mg per cent as the [illegible]

[illegible] ectively the phosphorus during the period of suboptimal insulin therapy (period 4 FIGURE 2) were 263 mg per cent and 4 98 mg per cent respectively In fact a close inspection of the mean daily levels of fasting blood sugar and plasma inorganic phosphorus for all periods in both FIGURES 1 and 2 suggest with a few exceptions described below that there is an inverse relationship between these two variables irrespective of the type of treatment The glucose phosphorus relationships just described are similar to those reported previously in diabetes[9,25] They are of interest since they are the reverse of the well known short term relationship between glucose and phosphorus which has been observed in nondiabetics following the administration of glucose or insulin

[illegible]

in levels of plasma inorganic phosphorus (periods 2 and 3 FIGURE 1, and periods 1 and 2 FIGURE 2) The mean decreases in levels of plasma inorganic phosphorus in patients R E and A I were 0 76 and 0 56 mg per cent, respectively Discontinuation of glucagon resulted in a prompt return of plasma inorganic phosphorus to control levels (period 4, FIGURE 1, and

period 3, FIGURE 2) The administration of carbutamide did not alter th hypophosphatemic response to injections of glucagon in either patien (period 7, FIGURE 1, and period 6, FIGURE 2), even though, in one of these patients (A F), carbutamide virtually abolished the hyperglycemic response to glucagon administration The decreases during glucagon administration in the mean daily levels of serum inorganic phosphorus below the control levels in patients R E and A F were 1 02 mg and 0 63 mg per cent respectively As was the case with glucagon administration during period of optimal insulin therapy, discontinuation of glucagon resulted in a prompt return of serum inorganic phosphorus toward control levels It should be noted that in patient R E the fall in serum inorganic phosphorus induced by glucagon in periods 3 and 7 was not associated with any significant increase in peripheral glucose utilization as measured by changes in the $\frac{I I}{I}$ ratio

Urinary reducing corticosteroids The mean daily urinary excretion of reducing corticosteroids was within the normal range in both cases Glucagon administration produced a slight but significant rise in the mean daily excretion of urinary reducing corticosteroids in the case of patient R E when she was being treated with optimal doses of insulin (periods 2 and 3, FIGURE 1), but this did not occur when glucagon was again administered while the patient was receiving carbutamide instead of insulin (periods 6 and 7, FIGURE 1) Neither glucagon nor carbutamide had any effect on the level of excretion of urinary reducing corticosteroids in patient A F

Phosphorus, potassium and sodium balances Slight increases in the mean daily urinary excretion of phosphorus were observed when both patients R E and A F were given glucagon either while receiving optimal doses of insulin or carbutamide in periods 2, 3, 4, 6 7, and 8 in Table 1, and in periods 1, 2, 3, 5 6 and 7 in Table 2 The increases in inorganic phosphorus excretion induced by glucagon were highly significant ($p < 0.01$) in the case of patient R E (only when carbutamide was given) but were not statistically significant ($p > 0.05$) in the case of patient A F Although the daily balances for sodium and potassium varied markedly from day to day and period to period in both patients, no relationship could be established between the changes in balances of sodium and potassium and the administration of glucagon and carbutamide either singly or together

Metabolic Effects of Sulfonylureas in Patients with Severe, Long-Standing, Unstable Diabetes of the Juvenile Type

Tolbutamide The oral administration of 2 0 gm of tolbutamide daily to patient E R (FIGURE 3) for 12 days did not lower the levels of blood and urinary sugar (periods 1 and 2) In fact, the glycosuria tended to increase while the patient was receiving tolbutamide It should be pointed out that this patient was receiving only sufficient insulin daily to maintain the fasting blood sugar at slightly above the normal range, and the mean 24 hour excretion of glucose in the urine at approximately 25 gm Ketonuria was

taneous fall in levels of blood and urinary sugar (period 3) the nitrogen

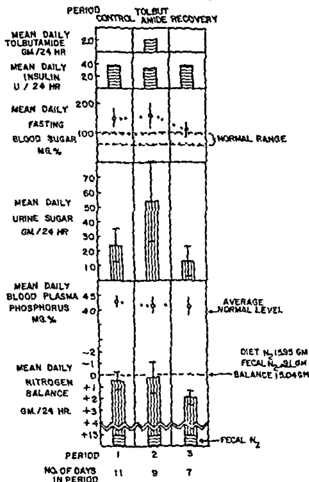


Figure 3 Effect of continued administration of tolbutamide in a severe unstable diabetic maintained on suboptimal daily doses of insulin. All data are expressed as mean daily values ±2 S.E. about the mean. Patient F. R., male, 50 years old, unstable diabetes, long duration.

balance became more positive even though the daily dose of insulin was

Chlorpropamide This compound was administered orally to patient E D during 2 different periods separated by an adequate recovery interval (FIGURE 4) On 24 U of insulin daily (20 U of lente insulin plus 4 U of crystal line insulin), the mean daily fasting blood sugar level during the 11 day control period was 247 mg per cent and the mean daily excretion of sugar in the urine was 50 2 gm (period 1) With the administration of 1 0 to 1 5 gm of chlorpropamide per day in addition to the 24 U of insulin per day for 16 days (period 2) the mean fasting daily blood sugar level was 188 mg per cent and the mean daily excretion of sugar was 26 7 gm The decrease in level of fasting blood sugar was just significant at the 5 per cent level while the decrease in the amount of glucose excreted in 24 hours was significant at less than the 5 per cent level With the discontinuation of chlorpropamide, both the mean daily fasting blood sugar and the mean 24 hour excretion of sugar returned toward control values (period 3) The mean daily fasting blood sugar level in period 3 was 234 mg per cent and the mean daily excretion of glucose 41 5 gm After a 10-day recovery interval chlorpropamide was administered again (period 4) On this occasion the daily dose of chlorpropamide was increased to 2 0 gm per day while the daily dose of lente insulin was decreased to 16 U per day, with the 4 U of crystalline insulin omitted There was no change in the 24 hour excretion of glucose

TABLE 3
PHOSPHORUS SODIUM AND POTASSIUM BALANCE DATA OF PATIENT F R MALE 50 YEARS OLD UNSTABLE DIABETES OF LONG DURATION*

	Period		
	1	2	3
No of days	10	10	6
Tolbutamide gm 24 hours		2 0	
Insulin U 24 hours	36	34	36
Urine inorganic phosphorus gm 24 hours	796† ± 048	794 ± 090	756 ± 106
Urine sodium mEq 24 hours	80 4† ± 13 6	96 5 ± 16 4	76 6 ± 16 4
Urine potassium mEq /24 hours	89 4† ± 27 2	88 4 ± 33 8	80 4 ± 21 2

* For further details see text and FIGURE 3 All urinary values are expressed as the mean ±2 S E about the mean

† To obtain the daily balance subtract the urinary value from the average daily intake urinary values less than the average daily intake indicate positive balance and urinary values greater than the average daily intake indicate negative balance

	Phosphorus gm	Potassium mEq	Sodium mEq
Sample daily diet	1 917	98 52	102 62
Average daily fecal excretion	−1 054	− 8 43	− 1 78
Average daily intake	863	90 09	100 84

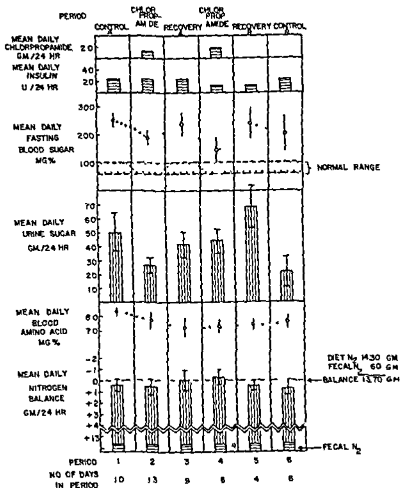


FIGURE 4 Effect of continued administration of chlorpropamide in a severe unstable diabetic maintained on suboptimal daily doses of insulin. All data are expressed as mean daily values ±2 S.E. about the mean. Patient F. D., female, 31 years old, unstable diabetes, long duration.

in the urine (43.9 gm in period 4 as compared to 41.5 gm in period 3) but the fasting blood sugar fell from a mean daily value of 234 mg per cent in period 3 to 142 mg per cent in period 4. The decrease of 92 mg in the mean daily fasting blood sugar level was significant beyond the 5 per cent level. When the chlorpropamide was discontinued but the daily dose of insulin maintained at 16 U of lente insulin per day, the fasting blood sugar level moved toward but did not exceed the control levels, whereas the mean daily

tion as measured by changes in the *A V* glucose difference This suggests that the phosphorus was not entering into the glycolytic cycle of peripheral tissues It is possible that, under the conditions described, the fall of inorganic phosphorus may represent entry of phosphorus in the glycogenic cycle in the liver, and that glucagon may be involved in the regulation of these processes A different type of study would be needed to establish or disprove the validity of this hypothesis

The failure of tolbutamide to lower blood or urinary sugar levels in patient E R is in agreement with the results of other investigators who have been unable to demonstrate a hypoglycemic action of tolbutamide or carbutamide in severe, unstable diabetics [15, 17] For this reason the lowering of blood and urinary sugar levels in patient E D , which appeared to be associated with the administration of chlorpropamide, is of interest The results suggest the possibility that some types of sulfonylurea compounds may have a hypo

glycemic response to chlorpropamide on the basis of stimulation of the insulogenic function of the pancreas It is reasonable to suspect that a different type of mechanism may be involved Chlorpropamide did not affect the daily nitrogen balances in this patient However we do not consider this as strong evidence against the possibility of an insulinlike' action of the sulfonylureas because the patient remained in nitrogen equilibrium while she was being suboptimally regulated with insulin without chlorpropamide

The findings that have been discussed are not all readily explainable on the basis of a single locus of action of sulfonylureas As pointed out, the probable hypoglycemic action of chlorpropamide in patient E D , who was presumed to be critically deficient in endogenous insulin, and the inhibition of the hyperglycemic response to glucagon by carbutamide in patient A F are difficult to explain on the basis of stimulation of the beta cells by the sulfonylureas to produce more insulin Nevertheless, the results of this study are not inconsistent with the hypothesis that the main action of sulfonylurea compounds is in the pancreatic islets This does not necessarily mean however, that the sulfonylureas stimulate the beta cells to release or secrete more insulin as some investigators have suggested [31, 34] It seems possible, for example, that the sulfonylureas might act by decreasing the ratio of glucagon to insulin through an inhibiting action on glucagon producing cells However, a hepatic effect of glucagon cannot yet be excluded on the basis of the work reported above

...ording to available evidence, the hypoglycemic action of sulfonylureas

of glucose

increasing the permeability of peripheral tissues for sugar, the findings described have been interpreted by some investigators as evidence that the hypoglycemic action of sulfonylureas is not insulinlike and, therefore, not the result of stimulation of beta cells by sulfonylureas [35, 37] This conclusion

seems reasonable to us but some investigators[8] hold that the sulfonylureas stimulate the beta cells to release or secrete small quantities of insulin directly into the portal venous system This small amount of released endogenous

peripheral glucose uptake as measured by available techniques It should be pointed out, however, that a direct action of insulin on the liver has not yet been unequivocally established[33] It appears to us that a more plausible

Glucagon is known to have a well defined action on the liver, but as yet no action has been established for peripheral tissues

One of the chief arguments raised to refute the possible action of sulfonyl urea compounds on glucagon production is that the compounds have no hypoglycemic effect in alloxan diabetic animals[34] in which presumably, the alpha cells are intact This may not be a sufficient basis for excluding the hypothesis of an alteration in the glucagon insulin ratio as a mechanism of action of sulfonylureas however it may well be that some insulin must be present in order that the effects of a diminution in glucagon may be observed This idea seems rational because, in the absence of insulin, the utilization of glucose by the periphery is too small to cause a lowering of blood sugar sufficient to expose the failure of glucagon to act

Summary and Conclusions

The metabolic effects of prolonged administration of carbutamide, tolbut amide, and chlorpropamide have been investigated in four selected diabetic subjects under controlled conditions in the metabolism ward of the Strong Memorial Hospital

Except for an effect on blood and urinary sugar levels, none of the com pounds that were studied appeared to have any direct or consistent effect on the following metabolic parameters (1) daily balances of phosphorus potassium, and sodium (2) daily balances of nitrogen, (3) daily levels of fasting plasma α amino acid nitrogen, and (4) daily urinary excretion of corticosteroids

The effects of sulfonylureas on hepatic gluconeogenesis and glycogenolysis were investigated in two diabetic subjects who showed a striking hypoglyce mic response to carbutamide This was done by studying the effect of prolonged administration of carbutamide on the ability of repeated daily intramuscular injections of glucagon to increase the urinary excretion of nitrogen as well as to increase the levels of glucose in blood and urine The influence of carbutamide on the ability of glucagon to depress the level of serum inorganic phosphorus was also investigated Previous studies from this laboratory had demonstrated that the increased protein catabolism that is an integral part of the hyperglycemic glycogenolytic action of glucagon is related to an enhanced hepatic gluconeogenesis from protein

Carbutamide markedly diminished the hyperglycemic and glycosuric response to glucagon in one patient but had no effect on this response in the other patient even though the drug had produced markedly decreased levels of blood and urinary sugar in both patients However, in neither patient was there any influence of carbutamide on the increased protein catabolism or the depression of serum phosphorus caused by the administration of glucagon These results suggest that the well established action of sulfonylureas in reducing hepatic gluco e output is not dependent upon either an inhibition of the hyperglycemic glycogenolytic action of glucagon or a selective interference with hepatic gluconeogenesis from protein

The present studies confirm the previous reports from this laboratory of the occurrence of an inverse relationship between the levels of fasting blood [illegible] The possible role of

[illegible] administration of tolbutamide and chlorpropamide to reduce the levels of blood and urinary glucose in two cases of severe unstable diabetes where there was presumed to be a critical deficiency of insulin Both patients were maintained on sub optimal doses of insulin One patient was given tolbutamide and the other chlorpropamide Tolbutamide was ineffective in reducing the levels of glucose in blood or urine chlorpropamide administration was associated with significant lowering of levels of blood and urinary sugar The findings suggest that the hypoglycemic action of sulfonylureas may not be entirely dependent upon remaining islet cell reserve and that the capacity may vary with different compounds

The metabolic effects of sulfonylureas that have been described are inconsistent with the hypothesis of a single mechanism of action such as a stimulation of the beta cells to secrete or release insulin However the results do not preclude the hypothesis that the central locus of action of sulfonylureas is pancreatic islet tissue It is possible that the action of sulfonylureas in reducing hepatic glucose output apparently without increasing peripheral glucose utilization is due to a reduction in the ratio of secreted glucagon to secreted insulin rather than to stimulation of the beta cells This possibility is analyzed

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METABOLIC STUDIES OF CHLORPROPAMIDE IN NORMAL MEN AND IN DIABETIC SUBJECTS*

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We have conducted short term and prolonged studies in normal men and in diabetic subjects to compare the physiological properties of chlorpropamide and tolbutamide. The primary objectives of the investigation were: first, to correlate the hypoglycemic effect with the serum sulfonylurea level for each compound; and, second, to compare the rates of disappearance of the two drugs from the blood. In addition, we performed metabolic balance studies in two normal men during the administration of chlorpropamide.

Methods

Serum sulfonylurea determinations. Serum concentrations of both chlorpropamide and tolbutamide were determined by the same analytical method. This method, developed by Toolan and Wagner for chlorpropamide and described elsewhere in this monograph, was found to serve equally well for the determination of tolbutamide.

The performance of the method in this laboratory is summarized in TABLE 1.

TABLE 1
PERFORMANCE OF THE ANALYTIC METHOD IN THE DETERMINATION OF SERUM CHLORPROPAMIDE AND TOLBUTAMIDE CONCENTRATIONS

Recovery of the sulfonylureas from serum	
Chlorpropamide	97.0 ± 1.2%
Tolbutamide	99.0 ± 0.8%
Features of the method	
Reproducibility (standards)	
±0.25 mg/100 ml sulfonylurea	
Precision (intraset serum replicates)	
±0.13 mg/100 ml sulfonylurea	
Accuracy (interset serum duplicates)	
±0.23 mg/100 ml sulfonylurea	

[illegible] mg/100 ml scattered among 13 sets

The recovery of both sulfonylureas added to serum was essentially complete

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The method, which showed excellent reproducibility, allowed a precision of approximately 0.1 mg per cent and an accuracy of approximately 0.25 mg per cent in serum measurements of sulfonylurea.

Additional data relevant to the application of the serum sulfonylurea method are provided in TABLE 2. Chlorpropamide and tolbutamide, as well as tolbutamide and the tolbutamide metabolite, are interrelated in terms of their relative absorbance intensity in the method. A low serum "blank" value was found.

TABLE 2
DATA RELEVANT TO THE APPLICATION OF THE SERUM SULFONYLUREA ANALYTICAL METHOD

Comparative quantitation data

	Relative absorbancy (on a weight basis)	
Chlorpropamide	100.0	—
Tolbutamide	83.8	100.0
Tolbutamide metabolite	—	38.5

Serum blank values (55 different serum samples from 5 normal, 2 diabetic, and 2 other subjects run in 22 sets)
0.22 ± 0.54 mg/100 ml (as chlorpropamide)

Method not corrupted by
(1) Products from hemolysis
(2) Existence of the tolbutamide metabolite

Mean ± standard deviation given for serum blank values.

The method is not corrupted by the products from hemolysis. Aliquots of 2 different blood samples devoid of sulfonylurea were briskly beaten with wooden applicators while the blood was clotting. The resultant deeply pigmented sera, when compared in analysis with nonhemolyzed "duplicates," were found to have increased in apparent sulfonylurea only by about 0.4 mg per cent. The degree of hemolysis produced deliberately in this evaluation was approximately 10 times greater than any experienced throughout the investigation being described. Mention is made of this observation concerning hemolysis, since plasma and serum tolbutamide methods in current use have a demonstrated[1] or likely[2-4] sensitivity to corruption by the products from hemolysis.

As an important consideration, the sulfonylurea method used in our studies is not corrupted by the existence of the tolbutamide metabolite[5] (1-butyl-3-p-carboxyphenylsulfonylurea). The metabolic half life of the tolbutamide metabolite was determined by techniques similar to those described below for tolbutamide. It was found to be approximately 9 times (8.6) shorter than the metabolic half life of tolbutamide. The metabolite thus disappears from blood approximately 9 times faster than tolbutamide and exhibits absorbancy approximately 2½ times less than tolbutamide in this method (TABLE 2). It is unlikely, therefore, that measured serum sulfonylurea levels, after tolbutamide administration, could be more than about 4 per cent in error because of the presence of the metabolite.

Other determinations. Fasting blood glucose, serum sodium, potassium, and chloride, and urinary nitrogen, creatinine, sodium, potassium, chloride,

17-hydroxycorticoids, and 17-ketosteroids were determined by methods previously used in this laboratory [6]

Subjects Two normal and 2 diabetic subjects were admitted to the Metabolism Research Laboratory and maintained on constant diets The 2 normal subjects (R R, male, age 23, 70 kg, and J D, male, age 20, 69 kg) received a diet that provided 200 mEq sodium, 113 mEq potassium, 17 3 gm nitrogen, 320 gm carbohydrate, and 2900 calories per day One diabetic patient (A R, female, age 62, 87 kg) received 178 gm carbohydrate and 2150 calories per day the other (R B, female, age 63, 73 kg) received 150 gm carbohydrate and 1400 calories per day

Eleven diabetic patients, chosen because they responded poorly to tolbutamide, were studied in the Endocrinology and Metabolism Out Patient Clinic of the University Medical Center

Results

The disappearance of chlorpropamide and tolbutamide from the blood Measurements of chlorpropamide or tolbutamide disappearance from blood were made after intravenous administration of each compound The series included observations on 2 normal and 2 diabetic subjects After an over night fast each subject received chlorpropamide or tolbutamide sodium* (30 mg/kg) at 8 00 A M, and food was withheld for 6 hours

Data for one of the normal subjects are presented in FIGURE 1 It is seen that the chlorpropamide serum concentration declined from about 21 mg per cent at 15 min to about 18 mg per cent at 6 hours whereas the tolbut amide serum concentration fell more rapidly from about 20 mg per cent at 15 min to about 8 mg per cent at 6 hours The serum chlorpropamide

amide Thus under the circumstances of this experiment, tolbutamide dis appeared from the blood approximately 8 times faster than chlorpropamide

It is of interest to note (FIGURE 1) that there was no significant difference between the [illegible]

amide As would be anticipated, there was no correlation between the serum drug levels and the blood glucose levels in these acute (6-hour) experi ments It is likely that under these conditions hypoglycemia was maximally induced when the serum sulfonylurea levels (after the administration of either drug) were above 20 mg per cent for 15 min, 18 mg per cent for 30 min or 17 mg per cent for 45 min Reversal of the progressive hypo glycemia at about 45 min was undoubtedly the result of effective counter action through physiological mechanisms that function to normalize the concentration of blood glucose

FIGURE 2 shows data for one of the diabetic subjects This subject had

* Chlorpropamide was supplied by D Iezzoni Chas Pfizer & Co Inc Tolbutamide was supplied by C J O Donovan The Upjohn Co, Kalamazoo, Mich

FIGURE 1 Serum sulfonylurea and fasting blood glucose levels after the intravenous administration of chlorpropamide and tolbutamide to a normal subject (J D, male)

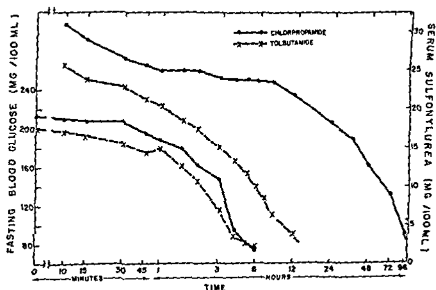


FIGURE 2 Serum sulfonylurea and fasting blood glucose levels after the intravenous administration of chlorpropamide and tolbutamide to a diabetic subject (A R female)

somewhat higher serum sulfonylurea levels during the study The higher serum levels probably resulted from the fact that the dose administered was based upon gross body weight and also from the fact that this subject was moderately obese Another notable characteristic of the sulfonylurea disappearance curves in this case is the marked difference between the chlorpropamide and tolbutamide serum levels Since the disappearance rates

In spite of the higher serum levels and the initial differential between the curves the disappearance rates for chlorpropamide and tolbutamide in this diabetic subject were quite similar to those for the previous normal subject (FIGURE 1) It should be noted that this diabetic subject as well as the other one in the series was previously treated with both sulfonylurea compounds The administration of chlorpropamide to both diabetic subjects was discontinued 9 days before these studies began

In the diabetic subject (FIGURE 2) there were almost identical progressive reductions in blood sugar after intravenous administration of equal quantities of the 2 drugs It is impressive that these acute blood glucose responses were remarkably similar in spite of the great difference that had developed in the serum sulfonylurea concentrations by 6 hours These results are consistent with those obtained in the normal subject and they suggest that the pattern for the blood glucose response was determined at an early time when the serum sulfonylurea concentration for each compound was sufficiently high to induce a maximal response

In the other 2 subjects the second normal and the second diabetic there was also no significant difference in the acute blood glucose responses after intravenous administration of the indicated amounts of chlorpropamide

evaluation is illustrated in FIGURE 3 for one of the diabetic subjects The disappearance rate is expressed as metabolic half life ($t_{1/2}$) which conventionally is defined as the time required for the serum concentration to decline to one half of a previous level

It is seen that after chlorpropamide administration at zero time there occurs a very rapid decrease in the serum concentration during the first 10 min (first 2 dots at 5 and 10 min) A rapid decline in serum concentration follows so that the metabolic half life of chlorpropamide during the time interval from 15 through 45 min is calculated to be 2 8 hours At about the first hour the serum concentration of chlorpropamide stabilizes and remains essentially unchanged up to the sixth hour During this 5 hour period the half life of chlorpropamide is described of course as infinite After this plateau phase the half life time fluctuates considerably until about 36 hours when a terminal steady state disappearance begins During

this phase of terminal steady state disappearance (36 to 144 hours) chlo *propamide is calculated to have a metabolic half life of 35 to 36 hours*

The disappearance of tolbutamide (FIGURE 3), after its intravenous adm istration can be described also in rate phases The three *indicated phase* for tolbutamide represent half life times of 3 3, 4 9, and 3 8 hours

The phases which have been described in this subject for chlorpropamid *and tolbutamide by half life* calculations were also observed in the secon diabetic and in both normal subjects The simplest interpretation whic can account for the phases observed after intravenous administration of eac compound is (1) an early (0 to 60 min) unstable space distribution, (2) redistribution (1 to 36 hours for chlorpropamide, 1 to 6 hours for tolbut amide) and (3) a stable space distribution (> *36 hours for chlorpropamide*

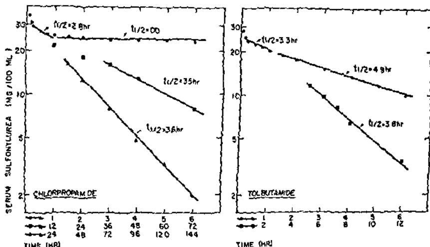


FIGURE 3 Metabolic half life times for chlorpropamide and tolbutamide in a diabetic subject (A R female)

> 6 hours for tolbutamide) It would be extremely interesting to make space distribution calculations for strategic timings in such an experimental series as this Since our studies did not extend to drug balance (excretory studies) this was not possible for these subjects It was interesting, however, *to make initial or zero-time* calculations of space distribution in a comparison of the 2 drugs Extrapolation from the 5 and 10 min points, in the 2 diabetic subjects gave chlorpropamide space volumes of 7 6 and 6 9 l and tolbutamide space volumes of 9 0 and 9 3 l *The mean chlorprop* amide and tolbutamide spaces in these 2 diabetics were 9 and 11 *per cent of body weight, respectively*

The difference between mean values of 7 3 and 9 2 l for chlorpropamide and tolbutamide spaces respectively would seem highly significant in the e 2 subjects From preliminary studies of deliberate blood sample hemolysis after chlorpropamide and tolbutamide administration it appears that the different *space distributions* of these 2 drugs may be explainable, to a major

extent on the basis of different concentrations in red cell water. The calculated zero time distribution volumes are probably smaller than the equilibrium distribution volumes and are discussed here only because they point up a difference between the 2 drugs. On the basis of published data by Wick *et al*[7] for tolbutamide in the eviscerated and nephrectomized rabbit, the equilibrium tolbutamide space might be anticipated as 13 to 19 per cent of body weight in man. Zero time distribution calculations have not been made for the 2 normal subjects, since 5- and 10 min. points on the sulfonylurea disappearance curves were not obtained.

TABLE 3
METABOLIC HALF LIFE MEASUREMENTS FOR CHLORPROPAMIDE AND TOLBUTAMIDE IN TWO NORMAL AND TWO DIABETIC SUBJECTS

Phase	Time interval (hours)	Metabolic half life time (hours): Normal subject: J.D.	R.R.	Mean ± M.D.	Diabetic subject: V.R.	R.B.	Mean ± M.D.
		Chlorpropamide					
A	<1	3.0	2.9	3.0 ± 0.1	2.8	2.8	2.8 ± 0.0
B	1 to 6	∞	∞	∞	∞	∞	∞
C	36 to 72	36	33	35 ± 1	35	37	35 ± 1
	72 to 120		—		36	37	
Postoral	24 to 120	35	34		32	—	
		Tolbutamide					
A	<1	1.8	1.8	1.8 ± 0.0	3.3	2.6	3.0 ± 0.4
B	1 to 6	5.1	6.1	5.6 ± 0.5	4.9	6.5	5.7 ± 0.8
C	6 to 12	4.3	4.8	4.6 ± 0.3	3.8	5.8	4.8 ± 1.0

Chlorpropamide tolbutamide disappearance ratio. Normal subjects 35/4.6 = 7.6, diabetic subjects 35/4.8 = 7.3

The disappearance data for all 4 subjects in the series are summarized in TABLE 3. A, B, and C represent the 3 phases previously described for intravenous chlorpropamide and tolbutamide in conjunction with FIGURE 3. There are probably no significant differences in the metabolic half life times between the 2 normal and the 2 diabetic subjects for any of the 3 phases with each of the drugs, with the possible exception of the tolbutamide phase A.

Half life times calculated after the cessation of oral (postoral) chlorpropamide administration are given in TABLE 3 for 3 of the 4 subjects as well.

The phase C and postoral metabolic half-life times, which represent terminal steady-state disappearance and a wide range of serum concentrations,

were used to arrive at a comparison of the rates of disappearance of these 2 drugs from the blood. This comparison is given as a ratio calculation below TABLE 3 and indicates that tolbutamide disappears from the blood from 7 to 8 times faster than chlorpropamide.

The relative potency of chlorpropamide and tolbutamide. It was clear that acute studies and classic dose response studies would yield information of limited value in a potency comparison of chlorpropamide and tolbutamide, especially if one attempted to apply such information in terms of long term oral therapy. On the other hand, it appeared as though meaningful data

FIGURE 4. Fasting blood glucose and fasting serum sulfonylurea levels in 2 normal men during the administration of chlorpropamide and tolbutamide.

could be derived from long term comparisons of the 2 drugs at various dosage levels and from a separation of the classic dose response study into 2 distinct components: the relation of dose to drug blood level and the relation of drug blood level to blood glucose response. Such an approach would take into account the facts that (1) the 2 drugs had markedly different half life times, and (2) the 2 drugs probably had different spaces of distribution. With these considerations in mind 2 normal men were studied for a period of about 4 weeks on an identical schedule of changing doses of oral chlorpropamide (FIGURE 4). During this period the men were carried through 2 cycles of symptomatic hypoglycemia. In a subsequent period they were observed during episodes of tolbutamide induced hypoglycemia. Much larger oral dosages of tolbutamide (at least 6 times larger) were required to produce comparable hypoglycemia and comparable serum sul-

fonylurea levels Daily measurements of the fasting blood glucose and the fasting serum sulfonylurea levels during oral administration of the 2 drugs revealed a consistent pattern

In FIGURE 5 the fasting serum sulfonylurea levels are plotted against changes in the fasting blood glucose level for the 2 normal men Standard

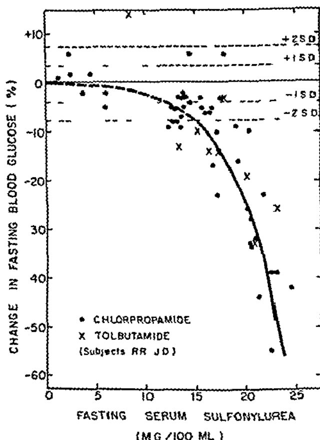


FIGURE 5 Correlation between change in fasting blood glucose and fasting serum sulfonylurea levels in 2 normal subjects

deviations relate to both men for daily fasting blood glucose measurements during an 8-day pretest period The curve was originally constructed in the absence of the tolbutamide points with reference only to the chlorpropamide points It is apparent however that when the tolbutamide points are included as they are in this figure they fit the previously constructed curve for chlorpropamide These results indicate that within the standardizations and extensiveness of these observations chlorpropamide

a decrease of 1 standard deviation in the fasting blood glucose in these normal men A decrease of twice the standard deviation in the fasting blood glucose resulted from a serum sulfonylurea level of about 15 mg per

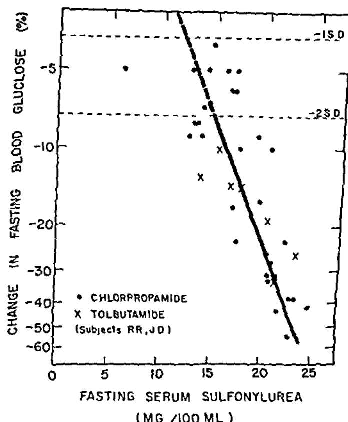


FIGURE 6 Correlation between change in fasting blood glucose and fasting serum sulfonylurea levels in 2 normal subjects (semilogarithmic presentation)

cent All sulfonylurea levels above 18 5 mg per cent produced decreases in the fasting blood glucose of at least twice the standard deviation Within the range of significant measurements, increments in serum sulfonylurea concentration of approximately 3 mg per cent double the effect on the fasting blood glucose

The data from FIGURE 5 are reproduced in FIGURE 6 with semilogarithmic coordinates Under this circumstance the relationship between the fasting serum sulfonylurea level and the change in the fasting blood glucose is represented by a straight line

Since chlorpropamide and tolbutamide on the basis of their serum concentrations are approximately equally active, and since tolbutamide disappears from the blood approximately 8 times faster than chlorpropamide it follows that chlorpropamide has the potentiality of being approximately 8 times more effective than tolbutamide on a weight basis, when the 2 drugs are used as oral therapeutic agents

It was of interest to obtain information on how the serum chlorpropamide concentration varies during a 24 hour period when 500 to 750 mg is administered each morning with breakfast (TABLE 4) Although the 2 subjects had

TABLE 4

VARIATION OF SERUM CHLORPROPAMIDE LEVELS DURING THE DAY AFTER SINGLE ORAL MORNING DOSES OF THE DRUG IN 2 NORMAL MEN

Subject	Day*	Chlorpropamide dose (gm/day)†	Time	Serum chlorpropamide (mg/100 ml)	Per cent of initial fasting level
R R (70 kg)	15	0.75	8 00 A M	15.7	100
			10 00 A M	23.5	150
			12 00 N	24.6	157
			2 00 P M	21.4	136
	16		8 00 A M	16.8	107
	20	0.50	8 00 A M	22.7	100
			3 00 P M	27.2	120
	21		8 00 A M	20.3	89
	21	0.50	8 00 A M	20.3	100
			5 00 P M	24.6	121
	22		8 00 A M	16.5	81
J D (69 kg)	15	0.75	8 00 A M	14.6	100
			10 00 A M	16.0	110
			12 00 N	18.0	123
			2 00 P M	17.0	116
	16		8 00 A M	15.8	108
	20	0.50	8 00 A M	24.6	100
			4 00 P M	26.8	109
	21		8 00 A M	20.6	84
	21	0.50	8 00 A M	20.6	100
			5 00 P M	21.2	103
	22		8 00 A M	18.2	88

* Days correspond with those in FIGURE 4

† Total dose taken shortly after 8 00 A M with breakfast

very similar fasting serum chlorpropamide concentrations on the same drug dosages they showed somewhat different but quite consistent variations in serum levels during the intervals studied The subjects developed maximum serum chlorpropamide concentrations of 123 and 157 per cent of the fasting levels about 4 hours after an oral dose at 8 00 A M The serum levels at 5 00 P M were 103 and 121 per cent of the morning fasting levels even when there was a moderate decline in the fasting serum level the following morning

Thus, a single dose of chlorpropamide at 8 00 A M would seem to provide elevated serum sulfonylurea levels (that is, relative to the fasting level) during the absorptive portion of the day (about 8 00 A M to 8 00 P M) and to allow the serum level to decline to a near fasting level early in the usually non absorptive portion (about 8 00 P M to 8 00 A M) of the day This feature, which is provided by the long metabolic half life time of chlorpropamide, may prove to be advantageous therapeutically

Clinical studies Eleven diabetic patients who responded poorly to tolbut amide were treated with chlorpropamide All responded better to chlor propamide at considerably lower daily dosages in most cases

The comparative response of the 11 diabetic patients to the 2 compounds is illustrated in TABLE 5 Each patient received each substance, at the indi cated dosages, for at least 3 days and usually for 1 week prior to the deter minations Complete comparative data were obtained for the first 3 patients

TABLE 5

COMPARATIVE RESPONSE OF PATIENTS WITH DIABETES MELLITUS TO ORAL TOLBUTAMIDE AND CHLORPROPAMIDE THERAPY

Patient	Tolbutamide			Chlorpropamide		
	Dosage (gm /day)	Fasting blood glucose (mg /100 ml)	Fasting serum tolbutamide (mg /100 ml)	Dosage (gm /day)	Fasting blood glucose (mg /100 ml)	Fasting serum chlorpropamide (mg /100 ml)
A R (95 kg)	3 0	191	5 3	0 75	138	14 2
E E (74 kg)	1 5	183	6 1	0 5	86	19 5
H H (60 kg)	3 0	176	4 6	1 0	88	29 3
R B (73 kg)	2 0	150	—	1 0	136	22 0
A C (58 kg)	2 0	210	—	2 0	146	—
F F (55 kg)	2 0	168	—	1 0	131	16 1
K H (72 kg)	1 0	155	—	0 5	83	22 2
S J (107 kg)	3 0	227	—	1 5	155	36 3
M R (98 kg)	1 5	153	—	0 5	109	10 2
P S (41 kg)	1 5	210	—	1 0	139	35 5
N M (75 kg)	1 0	131	—	0 25	99	7 5

listed The decreases in the fasting blood glucose levels experienced by these 3 patients on the indicated tolbutamide dosages were slight The relatively low fasting serum tolbutamide levels are to be noted In the same 3 patients considerably lower dosages of chlorpropamide produced higher serum levels of the drug and, concurrently, more intense decreases of the fasting blood glucose levels The same pattern of response described for the first 3 patients is characteristic of the other 8, as well

[illegible] are consistent with the [illegible] to its longer metabolic half life [illegible] e

schedule

TABLE 6

METABOL C BALA CE AND RENAL TXCRET ON STUD E N T O NORMA MEN DUR NG CHLORPROPAM DE ADM N TRAT ON*

Su	P d	D	F B S mg 00 m	U						
				Gm h		mEq 24 h			Mg 24 h ur	
				C	N g	a	K	C	CS	kS
R R Ag 23 0 kg	C Ch p p am C	8–0 8 9 33	88 8 9 — 0 90 9 8 9	2 2 06 30 2 2 98 2 3 0 8 2 6	4 9 3 3 0 0 3 4 3 3 3 8	9 8 8 86 04 6 0 6 20	8 34 08 8 04 8	93 63 2 0 9 80 02 6	0 0 8 8 2 0 2 8 6 2 9 3 4 0 0	5 0 6 22 9 5 0
J D Ag 20 69 kg	C Ch pr p am d C	8–0 8 29 33	82 9 85 45 86 8 8 89	2 3 2 2 4 3 5 2 0 2 6 09 2	2 3 4 6 4 2 0 0 4 9 2 6 0 3 0	80 6 93 83 24 8 66 3	03 0 08 60 6 96 8 22	0 86 2 8 202 4 266 3 8 8	8 0 8 6 9 5 2 3 5 6 2 5 8 6 5	0 8 8 5 2 8 5 20

D s re d w h h se RE 4
† Va pr n m an a d nge d d m na ns

An example of the difference in response to the 2 drugs is illustrated in FIGURE 7

Metabolic balance studies Two normal men were studied for a period of about 4 weeks on an identical schedule of changing doses of chlorpropamide The balance and renal excretion studies (TABLE 6) during chlorpropamide administration yielded results similar to those previously published from this laboratory for tolbutamide and carbutamide[6]

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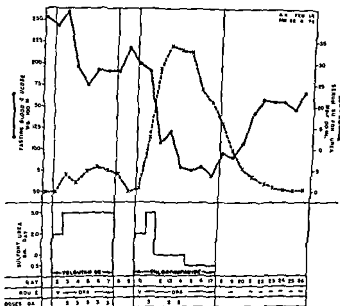


FIGURE 7 Fasting blood glucose and fasting serum sulfonylurea levels in a diabetic subject during the administration of tolbutamide and chlorpropamide

days when severe symptomatic hypoglycemia had been produced, urinary excretion of nitrogen and electrolytes was significantly decreased and the excretion of 17 hydroxycorticoids was increased Serum levels of sodium potassium, and chloride likewise were not affected

Summary and Conclusions

(1) A single analytical method has been found to perform well for the

od of normal men and diabetic

oes chlorpropamide

(4) Chlorpropamide, when compared with tolbutamide on a weight basis, exhibits a markedly greater clinical effectiveness

(5) The greater clinical efficacy observed with chlorpropamide seems adequately explainable on the basis of the capacity of the drug to produce and maintain higher serum levels on a given dosage schedule

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A COMPARISON OF THE ACUTE HYPOGLYCEMIC POTENCIES OF TOLBUTAMIDE AND CHLORPROPAMIDE*

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Comparisons of the clinical effectiveness of tolbutamide and chlorpropamide during their chronic administration to diabetic patients have indicated that smaller doses of chlorpropamide are required to produce a similar lowering of the blood glucose concentration At least part of this greater clinical potency of chlorpropamide as compared to tolbutamide is explained by its slower rate of disappearance from the body In the current studies the inherent hypoglycemic potencies of tolbutamide and chlorpropamide have been compared in terms of the acute decreases in blood glucose produced by the intravenous administration of equal doses of the two drugs to the same diabetic patients Use of the intravenous rather than the oral route of administration avoided possible differences in the rates of gastrointestinal absorption of the two compounds, and the acute nature of the study permitted a comparison of the effects of the drugs before the difference in their half lives had become manifest in significantly different plasma drug levels

Each patient was tested twice, once with tolbutamide† and on another occasion with an equal dose of chlorpropamide ‡ The patients were free of the influence of any hypoglycemic agent that is, they had received no long acting insulin for at least 48 hours, no regular insulin for 15 hours, no tolbutamide for 15 hours, and no chlorpropamide for at least 96 hours before any of the tests Preceding the tests the patients rested in bed after an overnight fast A 5 per cent aqueous solution of the sodium salt of the drug was injected into a peripheral vein within 1 min Blood samples were obtained through [illegible] at regular intervals after [illegible] blood glucose [illegible] opamide and tolbutamide by the method of Toolan and Wagner [illegible] statistical significance of differences between responses to the two drugs was assessed by analysis of the mean differences of paired observations

As shown in TABLE 1 equal doses of tolbutamide and chlorpropamide produced similar decreases in the blood glucose concentration during the 3 hours after drug administration The differences between these declines were not statistically significant The plasma concentrations of the drugs are shown in TABLE 2 Although at 2 and 3 hours the average tolbutamide levels were slightly less than the chlorpropamide levels, again the differences were not statistically significant

* [illegible] Diabetes Research Program of [illegible] earch Grant A 1317 from the [illegible] blic Health Service Bethesda [illegible] l Related Metabolic Diseases Foundation Inc, New York N Y

† Supplied by The Upjohn Company Kalamazoo Mich

‡ Supplied by Chas Pfizer & Co Inc Brooklyn N Y

TABLE 1

MEAN DECREASES IN BLOOD GLUCOSE CONCENTRATION PRODUCED BY THE INTRAVENOUS ADMINISTRATION OF EQUAL QUANTITIES OF SODIUM TOLBUTAMIDE (T) OR SODIUM CHLORPROPAMIDE (C) TO THE SAME DIABETIC PATIENTS

Dose of drug (gm)	No of subjects	Drug admin istered	Fasting blood sugar (mg /100 ml)	Blood glucose decrease (mg 100 ml) — Minutes after administration					
				30	60	90	120	150	180
1 0	8	T	187	11 5	20 2	32 8	41 1	—	55 4
		C	173	9 7	23 2	30 5	41 3	—	55 7
		T C*	—	1 8 ± 8 4	−3 0 + 4 7	2 3 + 5 4	−0 2 ± 6 2	—	−0 3 ± 5 7
0 5	4	T	213	—	15 0	17 8	27 7	—	48 0†
		C	206	13 5	22 0	32 0	38 2	—	59 5†
		T C*	—	—	7 0 ± 9 5	−14 2 + 7 0	−10 5 ± 7 8	—	−11 5†
0 25	3	T	134	11 3	19 3	29 7	30 6	—	26 5
		C	133	—	15 3	19 3	23 0	—	27 0
		T C*	—	—	4 0	10 4	7 6	—	−0 5
0 125	2	T	202	6 5	15 0	23 5	31 0	33 5	39 0
		C	202	4 0	—	15 0	20 5	27 0	38 0
		T C*	—	2 5	—	8 5	10 5	6 5	1 0

* Mean difference ± one standard error between blood glucose decreases after tolbutamide and decreases after chlorpropamide

† Data from only 2 of the 4 subjects

TABLE 2
MEAN PLASMA CONCENTRATIONS OF TOLBUTAMIDE (T) AND CHLORPROPAMIDE (C) AFTER INTRAVENOUS ADMINISTRATION OF EQUAL QUANTITIES OF THE SODIUM SALTS OF THE DRUGS TO THE SAME DIABETIC PATIENTS

Dose of drug (mg)	No of subjects	Drug administered	Concentration of drug in plasma (mg /100 ml)		
			Minutes after administration		
			60	120	180
1 0	8	T	9 9	7 8	6 4
		C	8 8	8 6	8 1
		T C*	1 1 + 1 2	−0 8 + 1 8	−1 7 + 1 1
0 5	4	T	5 8	4 5	3 8†
		C	7 2	5 0	4 8†
		T C*	1 4 ± 1 3	−0 5 ± 0 2	—
0 25	3	T	3 0	2 0	1 5
		C	2 5	2 4	1 9
		T C*	0 5	−0 4	−0 4
0 125	2	T	2 8‡	0 7	0 5
		C	1 2§	1 1	—
		T C*	1 6	−0 4	—

* Mean difference + one standard error between plasma concentrations of tolbutamide and chlorpropamide
† Only 2 values included
‡ One of samples obtained at 90 min
§ Both samples obtained at 90 min

Thus at similar blood levels the two drugs produced a similar fall in blood glucose during the first three hours after their intravenous administration to diabetic patients These data cannot be applied directly to the chronic administration of the drugs however, Knauff *et al*[3] found during long term administration of tolbutamide and chlorpropamide that the degree of fasting hypoglycemia was a function of the fasting sulfonylurea level and that the same relationship applied to both drugs These findings suggest ... for chlorpropamide as compared to tolbut... opamide rather than to any ... of the two drugs

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STUDIES WITH CHLORPROPAMIDE IN DIABETIC PATIENTS*

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The clinical trials of chlorpropamide reported here were begun in April 1958 our experience to date therefore extends over only 5 months The studies were initiated with the following four objectives in mind first to determine the effectiveness of the drug as a hypoglycemic agent in the management of diabetes second to observe serum levels of the compound in patients under control third to determine whether or not long term effectiveness could be predicted by the blood sugar response in a single dose test and finally to study the possible toxicity of the drug by performing serial liver and kidney function tests and hematological studies

Clinical Material and Methods

Seventy eight patients with proved diabetes selected primarily on the likelihood of responsiveness to sulfonylurea compounds have been given chlorpropamide to date The group consisted of 48 females and 30 males The ages of this group varied between 35 and 83 years and all but 10 were above 50 years of age The age at onset of diabetes varied from 23 to 77 years in 10 patients this had been below the age of 40 years The duration of diabetes varied from a few weeks to 21 years in 60 patients this was more than 1 year

In all cases but one the patients were hospitalized and maintained on weighed diets The total amount of sugar excreted in the urine was determined for each 24 hour period The blood sugar was determined frequently usually 3 times daily before breakfast at 11 A M and at 3 P M The majority of the patients were ambulatory The diets provided from 130 to 200 gm of carbohydrate daily, with most in the range of 150 to 170 gm The caloric content varied from 1000 to 2000 calories daily often this was low so as to favor weight reduction In all but 21 patients the insulin requirement had been established either by long term usage or during a brief period of regulation prior to trial of the oral hypoglycemic agent In the 57 patients whose insulin requirement was known this was 40 units or below in 47 patients and 20 units or below in 22 patients

Blood sugar determinations were carried out by the *Somogyi Nelson* procedure Most blood sugar levels before breakfast were determined on venous blood most other samples were capillary For the determination

* The work reported in this paper was supported in part by grants from Chas Pfizer & Co Inc Brooklyn N Y and The Upjohn Company Kalamazoo Mich
† Research Fellow

of the level of chlorpropamide we used the method of Spingler[1] as modified by R H Carmichael In this procedure serum is extracted with isoamyl acetate and treated with 1 fluoro 2-4 dinitrobenzene and the resulting color measured in the Beckman spectrophotometer at 345 mμ Prior to the beginning of therapy most patients had base line liver function studies including

function studies are being done after each 3 months of treatment with the drug

Sulfonylurea response test Prior to long term trial with chlorpropamide a single dose response test was done In general the procedure used was the same as that previously described in studies with carbutamide and tolbutamide[2] The test was carried out as follows (1) long acting insulin was omitted for 24 hours prior to the test day with crystalline insulin being substituted where necessary and (2) at 8 A M on the day of the test 1 0 gm of chlorpropamide was given orally after an overnight fast and glucose determinations were done on 3 blood samples (a) fasting before chlorpropamide (b) fasting 12 noon and (c) at 3 P M (about 2½ hours after a regular noon meal)

The maintenance dose in almost all patients was either 0 25 or 0 50 gm /day A few patients received 1 gm day for short periods during the early period of the study

Results

Response tests The response test was carried out in 71 patients In 37 a fall in blood sugar of 20 per cent or more occurred at both 4 and 7 hours
Fi
7

periods varying from a few days to 140 days

the dr for periods longer than one month and 48 for more than 10 days

reasons poor control on maintenance

nonmedical reasons 11

The average
test dose wer

Minor toxic effects consisted of minimal gastrointestinal

2 patients and a skin rash in another One patient developed jaundice after 3 weeks of therapy with 0 50 gm /day He was hospitalized, and initial laboratory studies showed the following results bilirubin, total, 3 9 and direct, 3 0 mg /100 ml , Bromsulphalein retention, 49 per cent at 45 min , alkaline phosphatase, 19 2 Bodansky units, thymol turbidity, 0 3 units, and cephalin and thymol flocculations negative The differential white blood cell count showed an eosinophilia of 10 per cent, falling to 1 per cent after 12 days Liver biopsy showed canalicular bile stasis, but no evidence of hepatocellular damage The patient improved with conservative therapy, and the last follow up 2 months later showed the following laboratory results bromsulfalein retention, 9 per cent in 45 min alkaline phosphatase, 4 7 Bodansky units thymol turbidity, 0 8 units, and thymol flocculation negative

Discussion

Of the 50 patients who showed a fall of more than 20 per cent in blood glucose in the 4 hour sample of the response test 41 achieved good control Of the 31 patients showing a fall of 20 per cent or more in both 4 and 7 hour samples, 26 achieved good control These results in conjunction with the finding of an adequate serum level of chlorpropamide at 4 hours suggest that the response test carried over 7 hours has no advantage over the 4 hour test Eighty per cent of patients having a satisfactory response with either test are likely to have their diabetes controlled Of the 13 patients who had less than a 20 per cent fall at 4 hours, 7 achieved good control, 2 fair control, and 4 poor control It thus seems that a poor response test does *not preclude* subsequent control of diabetes with chlorpropamide

The serum chlorpropamide determinations suggest that adequate levels are attained within 4 hours and maintained over 24 hours after an initial dose of 1 gm , and throughout longer periods with doses of 0 25 to 0 50 gm day

In evaluating the effectiveness of chlorpropamide in controlling diabetes, the role of diet must be considered As most of our patients were obese their diets were restricted in calories as well as carbohydrate content The average weight loss of our patients during this study period was 2 9 kg and in 11 patients chlorpropamide could be discontinued and control *maintained* with diet alone

The only serious toxic manifestation in our group of patients was the occurrence of jaundice in a 54 year old man The clinical picture, the laboratory data, and the histological appearance of a punch biopsy of the liver were all similar to findings in jaundice produced by chlorpromazine and methyl testosterone It is noteworthy, however, that our patient had a past and family history of jaundice, the nature of which was not determined

Summary and Conclusions

(1) Seventy-eight patients, selected mainly on the likelihood of responsiveness to sulfonylurea compounds were given chlorpropamide All but 10 were above 50 years of age, and the onset of diabetes occurred before the age of 40 in 10 patients

(2) In 71 patients the response of the blood sugar to 1 gm of chlorpropamide was determined at 4 and 7 hours Eighty per cent of the patients who had a fall of 20 per cent or more at 4 hours subsequently achieved good control of hypoglycemia and glycosuria with maintenance doses Of the 72 patients who received the drug for a period of up to 140 days 68 per cent achieved good control 15 per cent fair control, and 14 per cent poor control

(3) Serum chlorpropamide levels ranged between 3 and 32 mg per 100 ml, with an average of 10 mg per 100 ml

(4) One patient with a past and family history of jaundice developed an obstructive type of jaundice 3 weeks after onset of therapy There was complete recovery Two patients had minor gastrointestinal symptoms and 1 patient developed a skin rash

(5) These preliminary studies indicate that, in selected patients chlorpropamide is an effective hypoglycemic agent Wider experience with the drug is needed to evaluate its long term usefulness and incidence of untoward effects

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USE OF CHLORPROPAMIDE IN DIABETES MELLITUS IN USUAL DIABETIC PATIENTS AND IN PRIMARY AND SECONDARY TOLBUTAMIDE FAILURES*

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In a preliminary report[1] on management of diabetes with chlorpropamide (Diabinese†) successful control was noted in 58 per cent of 45 patients studied Further observations were made in 70 cases for periods up to 8 months We were particularly interested in observing the effectiveness of chlorpropamide in a group of patients who were primarily unresponsive to tolbutamide (within the first month of treatment) and in a larger series of cases classified as secondary tolbutamide failures that is, patients in whom diabetes had slipped out of control after a variable period of satisfactory management with tolbutamide

Method of Study

Diabetic patients who were taking insulin an occasional patient not controlled with diet alone and those not responding to tolbutamide were used in this investigation The first few patients were hospitalized, but later the drug was given to clinic and ambulatory private patients Diet was prescribed according to optimal caloric requirements

In the hospital, fractional urinalyses for sugar and acetone were made by the ward nurses before meals and at bedtime Each test showing more than a trace of sugar (Clinitest‡ modification of Benedict's test) was covered by an injection of regular insulin ranging in dosage from 5 to 20 U depending upon the percentage of sugar present In this way we were able to prevent ketosis and at the same time observe the degree of effectiveness of chlorpropamide Daily fasting blood sugar an occasional blood sugar after a meal, and 24 hour quantitative urine glucose determinations were made as added parameters of control[2]

[illegible]

3 days indicated that these requirements had been attained, patients were given a supply of medication and discharged for follow up in the outpatient clinic Particular stress was laid on the need for frequent urine tests at

* [illegible]

Inc Brooklyn N Y

‡ The Clinitest apparatus and tablets are manufactured by The Ames Co, Inc Elkhart Ind

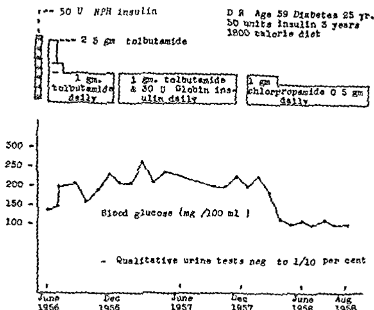


FIGURE 3 Good control with chlorpropamide after tolbutamide secondary failure

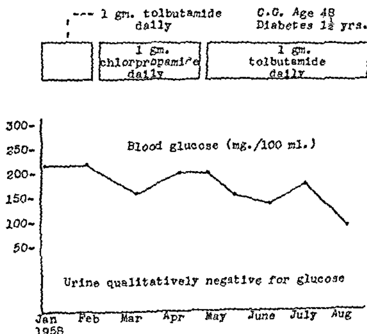


FIGURE 4 Good response to a second trial of tolbutamide after discontinuing chlorpropamide because of disagreeable taste

quate control, as measured by the criteria described above Six of the 28 patients who failed to respond to chlorpropamide were brittle type diabetics requiring large doses of insulin and having a pronounced tendency to acidosis For reasons such as fear of injections or recentness of onset of diabetes, 4 patients had not previously taken insulin These cases were easily controlled with chlorpropamide Three patients who had been taking insulin for more than 3 years remained well controlled after chlorpropamide was discontinued However, one of these (FIGURE 1) developed hyper glycemia without glycosuria after 2 months without medication This was relieved by a small daily dose of 0 25 gm chlorpropamide Only 2 of the 10 cases classified as primary tolbutamide failures responded well to chlorpropamide One of these is shown graphically in FIGURE 2 Our experience with secondary 'or late tolbutamide failures was much better Twenty of the 31 (64 per cent) patients in this group were managed satisfac torily with chlorpropamide A representative case is shown in FIGURE 3 Another noteworthy feature was a good response to a second course of tol butamide in 6 patients after chlorpropamide was stopped because of side effects (FIGURE 4) We suspect that the patients who responded to a second course of tolbutamide did so because of a renewed respect for adherence to diet

Toxicity

Side reactions to chlorpropamide are listed in TABLE 2 Gastrointestinal symptoms of anorexia nausea vomiting and abdominal discomfort were the most prominent Chest pain severe enough to warrant cardiovascular investigation was noted in 8 patients Electrocardiograms in all of these showed evidence of coronary artery disease and two of these later developed myocardial infarction Chlorpropamide was continued in 2 of these cases and the chest pain gradually subsided In fact 2 patients were given chlor propamide with good effect soon after suffering an acute myocardial infarct Another prominent complaint was weakness usually of legs arms and abdominal muscles Two of the 7 patients with this complaint had

TABLE 2
SIDE REACTIONS TO CHLORPROPAMIDE

	No of patients*
Anorexia nausea vomiting abdominal discomfort	9
Chest pain	8
Muscle weakness	7
[illegible]	4
[illegible]	4
[illegible]	3
[illegible]	1

* Of a total number of 70 patients treated

responded as far as diabetic control was concerned, but the symptom was so severe that it became necessary to discontinue the drug Serum potassium levels determined before and during treatment with chlorpropamide showed no diminution

Hypoglycemic symptoms (anxiety hunger sweating) were noted in 4 patients These never progressed to coma and disappeared promptly with administration of sweet drinks and reduced dosage



FIGURE 5 Maculopapular rash on face neck and arms following administration of chlorpropamide

Four patients with itching two with associated maculopapular rash of face arms and legs were noted One of these (FIGURE 5) was severe enough to warrant discontinuing medication, but the others responded to antihistamine preparations without discontinuing chlorpropamide

Three patients had an excellent response to a daily dose of 1 0 gm, but asked for a change of medication because of a disagreeable taste sensation This was de cribed as a sick, s eet taste, even in the lips " In retrospect we believe this could have been avoided by starting with a smaller daily dose of 0 5 gm

One case of leukopenia with a white blood cell count of 2500 was noted This was not permanent, when medication was discontinued, the count

promptly returned to normal The same patient complained of loss of hair after chlorpropamide had been discontinued Examination failed to substantiate this disorder

A depression of thyroid uptake of radioiodine was noted in 6 patients The range of lowering was 4 to 15 per cent There were no associated symptoms of hypothyroidism and this reaction was not considered of clinical significance

No unusual effects on kidneys or liver have been noted to date The longest period of observation has been 8 months

One patient died of coronary thrombosis after having been controlled with chlorpropamide for 4 months He was known to have malignant hypertension and his death was in no way attributable to the drug

Conclusions

Of 70 diabetics studied 60 per cent were controlled satisfactorily with chlorpropamide

The most easily controlled were middle aged patients with relatively

Toxicity to chlorpropamide is related partly to dosage Gastrointestinal symptoms chest pain and muscular weakness were noted in one third of the patients studied Occasionally these were relieved by reduction in dose of the drug

In most cases a smaller dose of chlorpropamide gave as good if not better control than tolbutamide

Unstable diabetics of any age are poor candidates for control with chlorpropamide

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A LABORATORY AND CLINICAL STUDY OF CHLORPROPAMIDE IN AMBULATORY DIABETICS

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Pharmacological studies have indicated that chlorpropamide is a potent hypoglycemic agent that is relatively nontoxic With a background of this knowledge clinical application of the drug was instituted in the Diabetes Clinic of the Long Island College Hospital *i* months ago Six elderly mild diabetics were given 1 tablet (500 mg) of chlorpropamide 3 times a day (a total of 1 5 gm) on the first day and 1 tablet twice daily (a total of 1 gm) thereafter Four of the patients complained of severe muscle weakness and dizziness leading to discontinuance of the drug This initial experience prompted a more detailed investigation of the pharmacological properties of chlorpropamide in humans since the experience obtained with other oral hypoglycemic agents did not seem to apply to this more potent agent

A method of estimating chlorpropamide as developed by the Research Department of Chas Pfizer & Co Inc Brooklyn N Y as described elsewhere in this monograph by Toolan and Wagner was employed to study the level of the drug in sera of patients following single doses as well as during long term administration of the medication The results of these investigations are the subject of the present work Clinical observations concerning the efficacy of the medication observed while treating ambulatory diabetics will be discussed elsewhere

Serum Levels Following Single Doses Given Orally or Intravenously

Serial estimations of the serum level of chlorpropamide as well as blood sugar were made at 20 min intervals for 4 hours after the administration of single doses of the medication TABLE 1 shows a summary of the serum drug levels found after a 0 5 gm dose was given orally and after an equivalent dose of the drug was given intravenously as the sodium salt The intravenous injections were accomplished over a 2 min period Both diabetic and non diabetic patients were studied Two facts are to be noted First the drug was well absorbed although somewhat slowly when given orally Two hours after oral administration high serum levels were obtained and within 4 hours the concentrations in serum were in a range similar to those obtained after the intravenous injection Second an appreciable quantity of the medication was still found in the serum 24 hours after its administration

These observations concerning the good absorption and prolonged retention of the drug in serum were again found when 1 gm doses of chlorpropamide were given orally and intravenously (TABLE 2 A and B) In this series oral administration was found to yield serum values at 4 and 24 hours that were higher than those obtained after a similar dose had been given intravenously

Thıs findıng suggested that slow absorptıon from the ıntestınal tract mıght be a factor causıng the prolonged elevatıon of drug levels ın the blood As can be seen ın TABLF 2 (C and D), after gıvıng sıngle oral doses of 1 5 and 2 gm, proportıonate ıncreases ın serum chlorpropamıde levels were not obtaıned ın 4 hours ın most experıments The larger doses led to hıgher concentratıons ın the serum after 24 hours

TABLE 1

SERUM LEVELS OF CHLORPROPAMIDE AFTER SINGLE DOSE OF 0 5 GM

Case	Dıabetıc Non dıabetıc*	Body habıtus†	Serum level (mg %) at 20 mın	40 mın	1 hour	2 hours	3 hours	4 hours	24 hours
A—Intravenous									
C H	N D	O	7 9	6 7	6 3	5 6	5 8	5 0	3 1
J S	N D	O	4 5	4 0	4 8	4 0	3 6	3 8	1 6
J (	N D	T	6 4	6 8	7 2	6 2	5 6	5 6	2 4
B S	N D	T	7 9	6 3	0	6 0	6 0	5 8	3 0
C H	D	M	7 2	7 0	5 7	6 4	5 7	2 2	—
D H	D	O	3 3	4 3	4 3	6 1	6 2	5 3	4 7
(R	D	O	7 2	4 5	4 3	—	5 9	4 3	0 3
D T	D	M	6 9	6 2	5 6	5 2	5 6	5 1	1 9
Q I	D	O	6 2	5 5	5 5	4 9	4 6	4 4	1 7
			Mean 6 39	5 70	5 63	5 55	5 44	4 61	2 34
			S D 1 55	1 17	1 06	0 81	0 83	1 11	1 30
B—Orally									
J Q	N D	O	2 4	2 6	3 0	7 (	6 3	5 8	—
T C	N D	O	0	0 9	1 7	2 5	3 3	3 6	2 5
C P	D	M	0	1 8	2 2	2 5	—	4 4	2 5
L F	D	M	0	1 0	1 4	4 8	5 7	5 0	2 0
			Mean 0 6	1 58	2 08	4 23	5 10	4 70	2 33
			S D 1 08	79	70	2 20	1 59	0 93	0 28

* Symbols D = dıabetıc N D = nondıabetıc
† Symbols O = obese T = thın M = medıum

The varıatıons ın blood levels obtaıned ın dıfferent patıents after the same dose of chlorpropamıde had been gıven (TABLES 1 and 2) could not be related to dıfferences ın absorptıon sınce ıt occurred after ıntravenous admınıstratıon as well The sıze of the patıent ınfluenced thıs to some extent In TABLE 2

of the drug assumıng that the medıcatıon remaıned ın the extracellular space In obese patıents the space was usually larger (25 to 27 per cent) and ın thın patıents such calculatıons found ıt usually smaller (16 to 18 per cent)

TABLE 2
SERUM LEVELS OF CHLORPROPAMIDE AFTER SINGLE DOSES

Case	Diabetic Nondiabetic*	Body habitus†	Serum level (mg. %) at 20 min.	40 min.	1 hour	2 hours	3 hours	4 hours	24 hours
A—Intravenous—1.0 gm.									
M V	ND	O	4.5	5.4	4.5	5.1	—	—	—
T F	ND	T	14.5	13.1	12.0	11.7	11.4	10.3	—
R A	ND	O	5.3	5.4	5.4	4.8	4.6	3.2	2.4
P C	D	T	6.4	13.7	10.5	10.3	9.8	9.8	3.6
W W	D	T	12.5	10.2	10.6	8.9	—	8.5	1.3
			Mean 8.6	9.6	8.6	8.2	8.6	8.0	2.4
B—Orally—1.0 gm.									
S B	ND	T	3.6	8.1	10.1	12.9	14.8	13.9	4.8
A M‡	ND	T	0	0	1.4	7.9	13.2	14.0	9.1
P M‡	D	O	0.2	6.1	8.9	13.0	15.1	12.1	9.4
T W	D	M	3.3	7.8	8.0	9.9	10.5	8.8	6.9
			Mean 1.8	5.5	7.1	10.9	13.4	12.2	7.6
C—Orally—1.5 gm.									
M G	ND	M	3.8	5.1	6.1	15.9	18.1	19.7	13.0
F G	ND	M	2.5	2.9	3.6	7.4	13.8	15.8	9.3
P D	D	T	1.0	5.2	8.4	14.5	12.3	13.2	4.4
M K	D	M	3.0	3.6					
			Mean 2.6	4.2	6.0	12.6	14.7	16.2	8.9
D—Orally—2.0 gm.									
S T	ND	T	—	—	22.4	—	—	21.4	16.7
J N	ND	M	4.4	8.4	9.2	21.5	15.6	15.2	15.8
A S	D	O	3.9	5.1	7.1	12.2	13.4	13.0	6.0
			Mean 4.2	6.8	13.4	16.9	14.5	16.5	12.8

* Symbols: D = diabetic; ND = nondiabetic.
† Symbols: O = obese; T = thin; M = medium.
‡ In 48 hours A. M. had a chlorpropamide level of 3.7 mg. % and P. M. 5.4 mg. %.

Serum Levels of Chlorpropamide After Continued Drug Administration

In view of the prolonged retention of the drug after a single dose, serum levels were studied in patients receiving continued daily doses. The amount prescribed was conservative in view of the previously mentioned experience in which 1 or 1.5 gm./day led to toxic symptoms. In general, 1 tablet of 500 gm. was given as a single daily dose. After 1 or 2 weeks the dosage was cut to 250 mg./day in older and very thin patients. In some instances, the latter type of patients were given 250 mg./day from the outset. Since one

of the important purposes of this investigation was to ascertain possible cumulative effects after continued administration, frequent changes in dosage were avoided

The above treatment was prescribed for 52 ambulatory diabetic patients who were given the medication from 6 to 26 weeks Serial estimations of chlorpropamide concentrations were made of the blood, which was also analyzed for glucose content and hepatic and renal function profiles The value of checking blood concentrations became apparent quite early Four

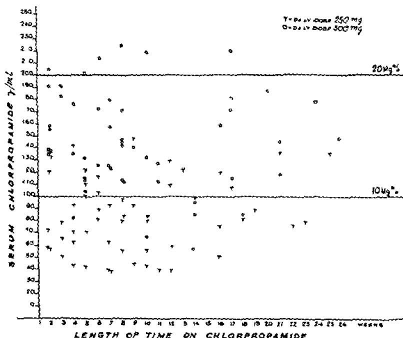


FIGURE 1 Serum chlorpropamide levels after prolonged administration

patients reported to the clinic regularly and insisted that they took the medication, yet zero levels of chlorpropamide were found in the blood Two confessed later that they had not, but the other two individuals insisted to the end that they had taken the medication Twenty two patients took 500 mg day, and 25 others took 250 mg day

In FIGURE 1 the blood levels of chlorpropamide are plotted against the length of time the drug was taken In the patients taking 250 mg, most points fall between 4 and 10 mg per cent This range is lower than that found in a control group of normal patients given the same dose, as reported elsewhere in this monograph by Carlozzi and Iezzoni The lower range might well be explained by patients' omission of occasional doses There were 2 instances in which the blood levels were consistently elevated, it is

possible that these patients took aspirin, which interferes with the chemical estimation of chlorpropamide

With a daily dose of 500 mg the scatter was much greater Again, 2 patients showed unexpectedly low values A history of omission of doses could not be obtained in these patients, but the trustworthiness of such a history cannot be guaranteed Serum chlorpropamide levels ranged from 11 to 22 mg per cent In general the lower values were found in the obese patients and higher values in small, thin patients, but this could not be correlated statistically

An observation constantly noted was that patients who had low serum drug levels on a given daily dose continued to have relatively low concentrations while receiving the same dose The same finding occurred in patients who showed high serum levels in the first 2 or 4 weeks, they continued to have the higher concentrations for months if the dose was left unchanged Thus, in the doses used, the blood level tended to be characteristic of the patient After a plateau level was attained a cumulative effect was not noted

Relationship Between Drug Levels and the Hypoglycemic Effect After Single Doses

Tests performed by estimating simultaneously the venous blood sugar levels and serum drug concentrations allowed the relationship between the 2 to be studied The former declined at a slow continuous rate while the latter rose gradually FIGURE 2 shows representative experiments in diabetics and nondiabetics After 0 5 gm was administered orally, the blood sugar began to fall when the chlorpropamide concentration was between 2 and 3 mg per cent (FIGURE 2 T C) The drop was maintained for 4 hours, when the drug levels were no higher than 6 mg per cent (FIGURE 2, L F) The hypoglycemic potency of chlorpropamide at these low dosage levels posed the question as to what effect larger doses might have When 2 0 gm were given orally and the drug was slowly absorbed (FIGURE 2, A S), the blood sugar also dropped slowly When absorption was rapid, leading to initial high serum levels, the drop in blood sugar was rapid (FIGURE 2, J T)

In practically all the oral experiments, the blood sugars continued to decline throughout the entire 4 hour period This was found in both nondiabetics and diabetic responders With the intravenous administration of the drug, however, nondiabetics showed a greater initial drop, after which the blood sugar tended to rise even when the drug blood levels were 10 and 12 mg per cent In some cases this rise in blood sugar in the face of an adequate drug level was sufficiently marked to lead one to suspect that drug concentration in the liver or other organs might be more significant than that in the systemic blood

Antidiabetic Action of Chlorpropamide

Many studies have reported an inability to correlate the single dose response of various oral hypoglycemic agents with their therapeutic efficacy The blood sugar, although important is in many respects only an indirect

indicator of the complex and subtle alterations of metabolism leading to the well known complications that occur in a diabetic organism Demonstration of the hypoglycemic activity of a drug does not necessarily denote that it is antidiabetic Even exogenous insulin was under suspicion by the late H Mosenthal who stated Insulin administered by injection, is not the complete remedy for diabetes, because the greater part of the injected insulin acts in the extrahepatic tissues and results in the deposit of glycogen in

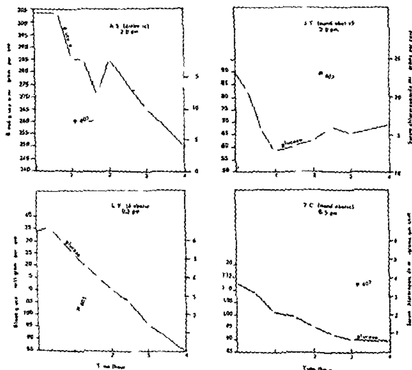


Figure 2 Blood sugar fall and chlorpropamide levels after a single dose of drug

them [4] In this light a total evaluation of the antidiabetic properties of chlorpropamide must await many years of clinical observation However, any attempt to initiate such an evaluation can begin by employing human diabetics of the maturity-onset type as test objects since such patients are known to respond to the drug

Thirty five of the 47 ambulatory patients who were selected for treatment with chlorpropamide were of this type and had previously been followed in the Diabetes Clinic for at least 6 months on other treatment The aim of treatment in this clinic is to obtain a physiological level of control and optimal nutrition The present group of patients was selected either because they

did not attain these criteria or because a comparison was desired between chlorpropamide and previous treatment The patients kept charts of their urine tests (Clinitest) before meals and frequent fasting blood sugars were obtained

The majority of our patients speak only Spanish, Italian or Arabic and live in families where the native dishes are chiefly of high carbohydrate content obesity is common In TABLE 3 (Group A) are assembled 15 uncomplicated obese mature diabetics of recent onset According to past observations of Duncan[1] and of this clinic[3] probably all could be excellently controlled by weight reduction with a resultant increase in glucose tolerance Varying degrees of lack of cooperation in dieting were the cause of poor diabetic control and all in Group A had not lost weight while presumably dieting In the first 4 cases the addition of chlorpropamide was definitely of value obtaining a level of control that was not obtainable in the previous period The fifth patient demonstrated that chlorpropamide does not allow unlimited eating of cake she refused insulin therapy

The next 7 patients had received insulin for at least 1 year prior to this study Replacement of the insulin with chlorpropamide led to better, or at least equivalent results in all but 1 patient (N O C) Many of these patients volunteered the information that they previously had frequent mild attacks of hunger that had forced them to eat more and that they felt better without insulin Despite these allegations weight loss was uncommon even when on chlorpropamide

The last 4 patients had previously received tolbutamide With the exception of patient N O C who did badly with all attempts at treatment 0 25 gm of chlorpropamide seemed to do as much as 1 gm of tolbutamide Control of these patients might have been better if larger doses of chlorpropamide had been used

In TABLE 3 Group B is composed of a similar type of obese adults with diabetes of long standing They had received insulin for 9 or more years and had remained obese The replacement of the insulin with chlorpropamide led to better control in half these patients Only 2 patients had previously been given tolbutamide chlorpropamide in 0 5 gm doses appeared to be slightly superior to 1 5 and 2 gm of the former drug

Diabetics who are thin pose a different problem Adequate nutrition must be given and thus from the onset their diets contain a relatively large amount of carbohydrate TABLE 4 shows the results with thin adult diabetics Group C contains 3 patients who were always thin developing diabetes later in life This number was small but seemed to show the value of the oral

before coming to the clinic usually due to neglect

serious illness or previously undiagnosed diabetes Insulin was usually prescribed when the patients first presented themselves to allow an ample diet to be given When chlorpropamide was given a gradual reduction of

TABLE 3
MATURITY ONSET TYPE OF DIABETES OBESE PATIENTS

Patient	Age	Duration (years)	Insulin (units) daily	Urine glucose (per cent)	Blood glucose (mg %)	Tolb * (gm) daily	Urine glucose (per cent)	Blood glucose (mg %)	P 607† (gm) daily	Urine glucose (per cent)	Blood glucose (mg %)
Group A relatively recent onset no weight loss											
P U G	51	1	0	0	186–238	—	—	—	0.5	0	116–140‡
M O L	40	1	0	0	162–197	—	—	—	0.25	0	96–130‡
P I A Z	42	1	0	1 2	158–213	—	—	—	0.25	0	96–120‡
N A D	68	8	0	0	200–266§	—	—	—	0.25	0	140–144
S A V	50	3	0	2	273–348§	—	—	—	0.5	0–¼	225–259§
I O R	61	1	20 PZI	0–2	144–155	—	—	—	0.5	0–¼	136–155
O N A	53	5	15 NPH	½–2	149–197	—	—	—	0.25	0	96–140‡
V I G	64	1	20 NPH	0	116–140‡	—	—	—	0.25	0	73–116‡
Z A R	67	3	15 NPH	0	160–202	—	—	—	0.25	0	102–138‡
M O N	71	2	25 NPH	0	171–191	—	—	—	0.25	0	181–184
K O I	65	5	38 NPH	0	181–202	—	—	—	0.25	0	165–176
N O C	65	4	35 NPH	0–¼	223–260§	3	0–¼	181–298§	0.75	0.1	202–266§
H A Y	46	6	0	0	244–250§	½	0	108–113‡	0.25	0	130–208
V I T	50	3	0	2	290–307§	1	0–¾	158–176	0.25	0	162–181
A Y I	71	7	0	0–2	156–180	1	0–2	158–181	0.25	0	178–197
Group B Obese at onset long standing diabetes still obese											
K A R	59	11	0	0	190–240	1 (BZ 55)	0	160–161	0.25	0	113–140‡
I O V	67	11	35 NPH	0–2	220–264§	2	0 tr	268–398§	0.5	0	176–197
D I I	67	16	25 NPH	0–2	260–318§	1.5	¼ ¾	360–396§	0.5	0	245–273§
I O N	56	11	35 Lente	0.1	162–298§	—	—	—	0.5	0	232–316§
M I I	61	9	12 NPH	0–2	197–232§	—	—	—	0.5	0–tr	113–148‡
S P F	57	21	20 PZI	0–2	218–219§	—	—	—	0.25	0	109–144‡

* Tolbutamide
† Chlorpropamide
‡ Excellent control urines always free of glucose fasting blood sugar 80 to 120 mg per cent at most times
§ Bad control urines usually contain sugar fasting blood sugar usually over 250 mg per cent
The maximum and minimum urine and blood sugars during the period of observation are given

TABLE 4
MATURITY ONSET TYPE OF DIABETES THIN PATIENTS

Patient	Age	Duration (years)	Insulin (units) daily	Urine glucose (per cent)	Blood glucose (mg %)	Tolb * (gm) daily	Urine glucose (per cent)	Blood glucose (mg %)	P 607† (gm) daily	Urine glucose (per cent)	Blood glucose (mg %)
Group C always thin											
Q U I	65	8	—	0	166–200	—	—	—	0 25	0	150–197
M A R	84	2	—	0 tr	186–200	0 5	0	166–200	0 25	0	144–158
C A S	48	1	—	—	—	2	0	104 124‡	0 25	0	120–132‡
Group D thin obese at onset recent diabetes											
F A V	71	4	65 NPH	0	122 136‡	—	—	—	20 NPH 0 5	0 ¼	167–202
K I I	68	2	35 PZI 20 PZI	0 1	148 200§	—	—	—	0 5	0–tr	137–161
M I R	65	3	15 reg	0	162 171	—	—	—	0 25	0	166–213
T O R	76	2	15 NPH	0 tr	132 181	—	—	—	0 5	¾–1	202–281§
Group I thin obese at onset long standing diabetes											
Z I S	64	14	24 NPH	0	96–180	—	—	—	0 5	0	124–165
S A D	73	18	25 PZI	0–1	213 334§	—	—	—	0 5	0	130–176
I I O	69	12	15 Lente	1–2	213–238§	—	—	—	0 5	0	166–184
I R A N	65	25	25 Globin	0–2	158–190§	—	—	—	0 5	1–2	195–232§
S C H	73	21	25 NPH	0	197–208	6	0–2	220–258§	10 NPH 0 25	0	166–171
S U N	69	21	0	0	140–160	1 5	0	136–162	0 5	0	102 106‡
B O D	74	16	0	0–¼	236–240§	—	—	—	0 25	0	225–232§

* Tolbutamide † Chlorpropamide ‡ Excellent control § Bad control

insulin was made in patients receiving 25 units or more The insulin was abruptly stopped in those taking less

It should be noted that the thin cases of recent onset (Group D) responded similarly to those of long standing diabetes (Group E) At the time that therapy with chlorpropamide was initiated, the level of control in these 11 patients (both groups) was excellent in only 1 patient and fair in only 2 even while on insulin Subsequently on oral therapy after discontinuing insulin 5 patients (K L E M I R, Z I S S A D, and F I O) of the 11 demonstrated diabetic control that was equivalent to or better than when on insulin Two cases who originally were badly controlled were markedly improved One patient (S C H) who was completely unresponsive to 6 gm tolbutamide day obtained fair control with 10 U NPH and 0 25 gm chlorpropamide The best result in this group was obtained in a cooperative patient (S U N) who attained a physiological level of control and gained weight with 0 5 gm chlorpropamide but could not do so with diet alone or with 1 5 gm tolbutamide This report represents a preliminary and therefore incomplete, study Out of the total of 35 patients in TABLES 3 and 4 there were 7 primary failures There is reason to believe that in many of our patients further manipulation of the dosage schedule might lead to better results

It is important to reemphasize clinical results in treatment The above charts giving urine and blood sugar level record only one of many criteria that should be used in assessing the antidiabetic quality of a medication In our obese diabetics the mere prescribing of a diet without adequate cooperation by the patient had failed and many had such complaints as pruritis vulva and thirst alleviated by the oral hypoglycemic agent In many thin adult patients chlorpropamide allowed better utilization of the ingested food leading to desired gains in weight Such results can only be inferred from charts but again should be mentioned

Toxicity

To study the incidence of toxicity the entire series of 47 ambulatory patients was reviewed It has already been stated that with a daily dosage of 1 5 gm 4 out of 6 patients had severe asthenia, muscular weakness and lightheadedness In the blood of one of these patients there was a drug level of 46 6 mg per cent and a blood sugar of 225 mg per cent after 2 weeks of medication Such incidences of toxicity must be attributed to overdosage rather than to the drug itself Recurrence of severe diabetic symptomatology occurred in 2 young adults with severe diabetes in whom the insulin was reduced when chlorpropamide was given The latter should be attributed to nonresponsiveness rather than toxicity Two patients without previous gastrointestinal complaints developed acute cholecystitis confirmed by surgery 1 and 2 months after taking chlorpropamide These were probably coincidental

In all 2 patients had pruritis while on therapeutic doses requiring discontinuance of the drug and 2 patients had dizziness and nausea One of the patients with pruritis previously had a pruritic rash when taking tolbut

amide (Orinase) All symptoms quickly disappeared with discontinuanc of the medication Serial hematological surveys on all patients have reveale no anemia a slight leukocytosis (10 to 11 thousand) with a normal differ ential leukocyte count was frequently encountered without adequate expla nation Serial liver profiles also did not change In this connection i should be mentioned that 3 diabetic patients with known hepatic cirrhosi have been given chlorpropamide Liver biopsies had established the diagno sis in all but 2 had normal liver function tests and only 1 was active a shown by a markedly elevated serum transaminase The latter patien returned to normal while on chlorpropamide therapy The liver profile ir the other 2 patients remained normal

The most serious manifestation of toxicity was encountered in a 56 year ok man who previously had urticaria when given Orinase Immediately after the intravenous administration of 1 gm chlorpropamide, he went into a state of shock that responded to an intramuscular injection of Chlor Trimeton Blood sugars prior to and during the episode were 167 and 165 mg per cent respectively

Summary

Chlorpropamide is an extremely potent hypoglycemic agent that is quickly absorbed and slowly excreted Oral administration in therapeutic doses leads to predictable drug levels in the serum It has demonstrated favorable antidiabetic activity during the period of study (6 months) and can effectively replace insulin in diabetics with maturity onset With it a better control can be obtained in that large segment of the diabetic population that does not fully cooperate in the dietary management Dosagewise it is more potent than Orinase Toxicity in therapeutic doses did not constitute a problem

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CLINICAL EXPERIENCE WITH CHLORPROPAMIDE*

Barnett Greenhouse

Grace New Haven Community Hospital New Haven Conn

medication for diabetes

Clinical Pilot Study

Chlorpropamide was subjected to a clinical trial in 50 nonhospitalized diabetic patients ranging in age from 18 to 87 with an onset age of 18 to 72 years and a duration of the disease from 1 day to 33 years Two thirds of the patients studied have been on insulin for various periods to 33 years, and one third of these were on dosages of 50 to 150 U

The patients were under close clinical observation and controlled by frequent laboratory tests designed to disclose any toxic effects on the liver, kidneys and blood forming organs Laboratory profiles including blood sugar urinalysis peripheral hematogram alkaline phosphatase thymol turbidity and flocculation and blood urea nitrogen were done at weekly biweekly and monthly intervals

The compound was used in three basic clinical situations (1) transfer from Orinase (2) conversion from insulin, and (3) in newly discovered or previously untreated cases

Transfer from Orinase to chlorpropamide In the initial test series Orinase controlled diabetics (twenty two patients) were shifted to chlorpropamide for comparative evaluation on a gram for gram basis an 0 5 gm tablet of chlorpropamide replacing an 0 5 gm Orinase tablet Control of the diabetes proved adequate but

chlorpropamide per day produc

changes in one third of the p

category of side effects of the hypoglycemic sulfas with varying degrees of intensity The subjective complaints were predominantly of a hypoglycemic nature with CNS circulatory and renal manifestations Other side effects noted were gastrointestinal disturbances and allergic skin reactions

There was no doubt that chlorpropamide lowered the blood sugar the response was prompt and dramatic exhibiting a greater potency than Orinase The side effects were always reversible and gradually subsided when

Better tolerance was achieved

osage although some patients

subsequently made available

* The investigation reported here was supported by a research grant from Chas Pfizer & Co Inc Brooklyn N Y

with a lowering of the daily dose level below 500 mg, there was a decided improvement in clinical usefulness Patients were successfully managed on the lower dosages with smooth control of hyperglycemia and glycosuria and with a low incidence of side reactions Transfers from Orinase to chlorpropamide were subsequently made by replacing a 500-mg Orinase tablet with a 200 mg tablet of chlorpropamide The medication was given in divided doses immediately after meals to minimize side reactions and to avoid gastric irritation

Chlorpropamide was also found to restore control where Orinase effect declined or failed on long use (six patients) One insulin patient who did not respond to Orinase responded readily to chlorpropamide and discontinued insulin

Conversion from insulin to chlorpropamide Generally where Orinase was known to be primarily ineffective chlorpropamide was also found to be

TABLE 1
DURATION OF CHLORPROPAMIDE THERAPY IN PATIENTS WITH DIABETES MELLITUS

Duration months	Number of cases			
	Total	Still on therapy	Lost to follow up	Therapy discontinued
1	8	—	—	8
2	8	3	—	5
3	10	8	—	2
4	2	2	—	—
5	5	2	—	3
6	1	1		—
7	7	5	—	2
8	9	8	—	1
Total	50	29		21

without effect in therapeutic dosages (six patients) Larger doses of chlorpropamide however tended to reduce the insulin dose in some resistant cases (four patients)

withdrawn
added every
day At the same time the insulin was raised 2 to 4 U daily as needed to combat glycosuria

TABLE 2
CLINICAL TOLERANCE FOR CHLORPROPAMIDE SIDE EFFECTS*

Hypoglycemia (14)

CNS (11) agitation blurred vision confusion dizziness depression dullness drowsiness fatigue flushing grogginess headache hunger incoordination irritability lack of well being lethargy loss of concentration malaise, nervousness restlessness sleeplessness unsteadiness weakness
Circulatory (2) congestive failure dyspnea palpitation
Renal (1) albuminuria dyspnea peripheral edema

Allergic Skin Reactions (4)

Erythema Multiforme (1) pruritis (2) urticaria (1)

Gastrointestinal disturbance (12)

Anorexia constipation diarrhea distention flatulence halitosis heartburn nausea vomiting

Alcohol intolerance (1)
Crystalluria ? (2)
Stippling of red blood cells (1)

* Figure in parentheses gives number of cases

Actually the dosages of insulin and chlorpropamide were chosen arbitrarily and the patient was kept on the same dose of chlorpropamide as long as it proved effective and caused a decline in the insulin requirement. Meanwhile without anticipating the effect of the drug, a close watch was kept on the insulin need to keep the condition from getting out of control or to prevent insulin shock. The regimen lent itself readily to outpatient and office management and none of the patients required hospitalization.

New and untreated diabetics. Previously untreated and uncomplicated mild cases (4 patients) and a recently discovered adult diabetic patient responded promptly to chlorpropamide. As a routine a 200 mg. tablet was given once or twice daily until the patient was sugar free, then he was maintained on 1 tablet every morning after breakfast. Thereafter a second

DISTRIBUTION OF PATIENTS AND RESULTS OF CHLORPROPAMIDE THERAPY IN 50 ADULT DIABETICS

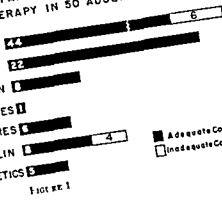


FIGURE 1

CORRELATION OF AGE AT TIME OF TREATMENT WITH RESPONSE TO CHLORPROPAMIDE

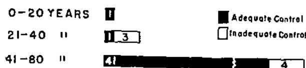


FIGURE 2

CORRELATION OF DURATION OF DIABETES WITH RESPONSE TO CHLORPROPAMIDE

AGE ONSET 1-20 YEARS (1 PATIENT)

< 1 YEAR 1

AGE ONSET 21-40 YEARS (12 PATIENTS)

< 1 YEAR 1
1-2 YEARS 1
3-5 " 1
6-10 " 1 1
11-20 " 1
> 20 " 5 1

AGE ONSET ABOVE 40 YEARS (37 PATIENTS)

< 1 YEAR 4 1
1-2 YEARS 4 1
3-5 " 10 3
6-10 " 7
11-20 " 3 2
> 20 1 1

Adequate Control
Inadequate Control

FIGURE 3

tablet was allowed after supper whenever the dinner test was positive for sugar

Toxicity Excluding toxic drug reactions inherent in overdosage, no persistent toxicity was encountered at therapeutic levels About the only evidence of toxicity, which relates to other sulfonamides as well, has been the development of some skin rashes and itching (four patients) Otherwise, there was no evidence of toxicity that could be determined by studies of liver function, renal function, and the blood elements No acetonuria was encountered in the responsive patients Dulling of the appetite was noted by some

CORRELATION OF DURATION OF DIABETES WITH RESPONSE TO CHLORPROPAMIDE

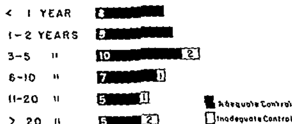


FIGURE 4

CORRELATION OF PRIOR INSULIN DOSE WITH RESPONSE TO CHLORPROPAMIDE

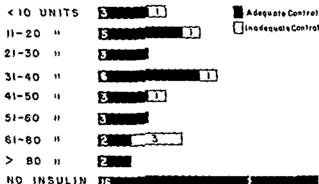


FIGURE 5

DAILY DOSAGE OF CHLORPROPAMIDE REQUIRED FOR MAINTENANCE CONTROL

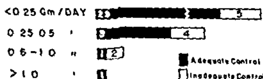


FIGURE 6

EXPERIENCE WITH CHLORPROPAMIDE
THERAPY DISCONTINUED

NO OF PATIENTS 50
UNDESIRABLE SIDE EFFECTS 6
SUGAR-FREE 9
INEFFECTIVE 6

FIGURE 7

Clinical Results One half of the group of 50 patients studied has been under continuous treatment for 3 to 8 months (TABLE 1) approximately one fourth of the patients were eliminated in the first 2 months because of *side effects (TABLE 2) or lack of response to chlorpropamide* Adequate control was obtained in 44 patients of whom 8 were maintained on chlorpropamide and insulin results were inadequate in 6 (FIGURE 1)

Chlorpropamide was found to be effective chiefly in patients more than

who have had diabetes for as long as 25 years (FIGURE 4) Patients on prior

tween 100 and 400 mg (FIGURE 6)

The drug was stopped in nine patients who became sugar free it was *ineffective in six patients and six patients abandoned medication because*

TABLE 3
CORRELATION OF DAILY DOSAGES OF CHLORPROPAMIDE WITH NUMBER OF SIDE EFFECTS

Side effect	Grams per day			
	<0 25	0 5	1 0	>1 0
Hypoglycemia and sequelae			14	
Gastrointestinal	1		11	
Skin		1	2	1
Alcohol intolerance		1		
Red blood cell stippling	1			
Crystalluria (?)	2			

Glycosuria and elevated blood sugars recurred after two to six months in three patients who had been on chlorpropamide therapy one of these was previously shifted to chlorpropamide becau e of secondary failure on Orinase

Comment

Preliminary and early use of chlorpropamide was undertaken without previous knowledge of the clinical effects of the drug During the exploratory phase guided by experience with Orinase chlorpropamide was first administered dose for dose to a selected group of Orinase controlled patients for comparative evaluation The response was prompt and unquestionable Chlorpropamide proved to be a potent and rapidly active hypoglycemic agent with an activity more than twice that of tolbutamide Do es in the magnitude of 1 0 gm were found to induce adver e side effects that for the most part were correlated with hypoglycemic manifestations and sequelae characteristic of the long acting insulins Expanding trials however showed chlorpropamide to be generally well tolerated and clinically effective in the therapeutic range of 100 to 400 mg and to have no significant undesirable by effects

In view of its enhanced potency and rapid action as observed clinically treatment with chlorpropamide was initiated without a priming dose In the newly treated diabetic the respon e to the drug was found to be rapid with a minimum dose from the start for conversion from insulin a ba ic dose

or relatively mild diabetics of the middle age or older group re pond best to the drug The responsiveness of the early case is very striking The mild diabetics and by that token the older patients are very sensitive to the drug

signific

propam

d sages tolerated beyond the therapeutic range and it may thus prove to be a truer mea ure of drug responsiveness than Orinase

A significant ob ervation involves the effectiveness of chlorpropamide in primary and secondary Orina e failure The development of a refractory tolerance to Orina e experienced on prolonged use of the drug is becoming evident al o with chlorpropamide Stopping medication at intervals to allow for a rest period and starting again when and if glycosuria recurs are mea ures aimed at preventing drug re istance

Chlorpropamide and Orinase have been used interchangeably and, currently a shift from one to the other also is being explored as a pos ible means of avoiding an acquired tolerance to either

Hypoglycemic reactions on small doses (200 mg) of chlorp e have been ob erved in sen itive individuals indicating the need f g t maintenance dose to the lowest level consistent with good nt

The management of diabetes with chlorpropamide is

of the marked blood sugar fluctuations and wide metabolic excursions characteristic of insulin Patients feel better and are less agitated, and excessive appetite is curbed by the anorexic effect of the drug, as well as by the normalization of the blood sugar Absence of acetonuria in the chlorpropamide-responsive patients reflects a favorable effect on carbohydrate metabolism with a stabilizing influence on the diabetic state See case reports below

Summary

Chlorpropamide (P 607 Diabinese), a new sulfonylurea derivative with a hypoglycemic activity similar to that of Orinase, but with enhanced potency and longer duration of action, is currently under clinical investigation

An early pilot study and expanding trials with this compound give promise of its therapeutic value in the treatment of diabetes It is proving to be an active and clinically effective oral hypoglycemic agent more than twice as active as tolbutamide In the therapeutic range of 100 to 400 mg, it appears to be safe, effective, and well tolerated, with minimal side reactions

Acknowledgment

I thank D G Iezzoni of the Clinical Research Department, Chas Pfizer Co, Inc, Brooklyn, N Y, for his interest and aid in the project reported in this article

CASE REPORT 1
Primary Orinase Failure

ORAL DIABETIC THERAPY – DIABINESE Conversion and Progress Report														Patient Name C N Chart No 1637
Date	Urine Sugar Test				Urine Keto Test				NPH Insulin				Dosage
1958	A M	A M	P M	P M	A M	A M	P M	P M	A M	A M	P M	P M	Per Day
Apr 22		0		0	BS*	150				20			500 mg
23		0		0						18			500
24		3		0						22			500
25		0		0						18			500
26		0		0						16			500
27		1		1						18			500
28		1		0						20			500
29		0		0						18			500
30		0		0						16			500
May 1		0		0						14			500
2		0		0						12			500
3		0		1						10			500
4		0		1						8			500
5		3		1						12			500
6		0		0						12			500
7		0		0						10			500
8		0		3						8			500
9		3		0						10			500
10		0		0						8			500
11		0		0						6			500
12		0		3						4			500
13		0		0						4			500
14		1		0	BS	138				6			500
Insulin discontinued on May 20, 1958													

* All blood sugars were performed at random by the Folin Malmros capillary blood sugar method and expressed in milligrams per 100 cc of blood

C N female age 48 weight 150 A diabetic on insulin for 22 years and currently on 35 U of NPH insulin and 1.0 gm of Orinase She was started on 250 mg Diabinese and 20 U NPH insulin daily and promptly became sugar free In 1 week she had an insulin reaction on one-half the previous insulin dose and gradually discontinued insulin

CASE REPORT 2
Secondary Failure on Orinase and Diabinese

ORAL DIABETIC THERAPY – DIABINESE Conversion and Progress Report														Patient Name R M Chart No 3538
Date	Urine Sugar Test				Urine Keto Test				NPH Insulin				Dosage
1958	A M	A M	P M	P M	A M	A M	P M	P M	A M	A M	P M	P M	Per Day
Apr 21		4		0	BS*	226				80			250 mg
22		4		1						80			750
23		4		0						80			750
24		1		0						80			750
25		4		0						80			750
26		4		0						80			750
27		0		2						80			750
28		0		0	BS	143				80			250
29		0		0						60			250
30		0		0						50			250
May 1		tr		0						45			250
2		0		tr						40			250
3		tr		0						30			250
4		0		4						30			250
5		0		–						25			250
6		1		0	BS	217				20			250
7		0		0						15			250
8		0		0						10			250
9		0		0						0			250
10		0		0						0			250
11		0		0						0			250

* All blood sugars were performed at *random* by the Folin Malmros capillary blood sugar method, and expressed in milligrams per 100 cc. of blood.

R M male age 46 weight 257 A diabetic for 4 years, he was originally converted to Orinase from 97 U of NPH insulin He continued sugar free on 1.0 gm Orinase daily for 7 months when his diabetes decompensated and he went back on insulin to 125 U daily His response to Diabinese was rapid and insulin was discontinued again Glycosuria recurred after 2 months and he was shifted to insulin and Orinase

CASE REPORT 3
Insulin Conversion

ORAL DIABETIC THERAPY – DIABINESE
Conversion and Progress Report

Patient Name: W. W.
Chart No. 3578

Date 1958	Urine Sugar Test A.M.	Urine Sugar Test A.M.	Urine Sugar Test P.M.	Urine Sugar Test P.M.	Urine Keto Test A.M.	Urine Keto Test A.M.	Urine Keto Test P.M.	Urine Keto Test P.M.	NPH Insulin U-40 A.M.	NPH Insulin U-40 A.M.	NPH Insulin U-40 P.M.	NPH Insulin U-40 P.M.	Dosage Per Day
May 30		0		0	BS*	216				20			200 mg
31		0		0						18			200
June 1		0		0						16			200
2		0		0						14			200
3		0		0						12			200
4		0		0						10			200
5		0		0						8			200
6		0		0						7			200
7		0		0						6			200
8		0		1						5			200
9		0		0						6			200
10		0		0						5			200
11		0		0	BS	158				0			200
	Insulin discontinued												

* All blood sugars were performed at random by the John Malmros capillary blood sugar method and expressed in milligrams per 100 cc. of blood.

W. W. male age 68 weight 174. A diabetic for 14 years patient was taking 20 to 30 U of NPH insulin. He was put on 20 units of NPH with 200 mg of Diabinese daily. He continued sugar free gradually reducing his insulin and discontinuing it in less than 2 weeks.

CASE REPORT 2
Secondary Failure on Orinase and Diabinese

ORAL DIABETIC THERAPY – DIABINESE
Conversion and Progress Report

Patient Name R M
Chart No 3538

Date 1958	Urine Sugar Test A M	A M	P M	P M	Urine Keto Test A M	A M	P M	P M	NPH Insulin A M	A M	P M	P M	Dosage Per Day
Apr 21		4		0	BS*	226				80			250 mg
22		4		1						80			750
23		4		0						80			750
24		1		0						80			750
25		4		0						80			750
26		4		0						80			750
27		0		2						80			750
28		0		0	BS	143				80			250
29		0		0						60			250
30		0		0						50			250
May 1		tr		0						45			250
2		0		tr						40			250
3		tr		0						30			250
4		0		4						30			250
5		0		–						25			250
6		1		0	BS	217				20			250
7		0		0						15			250
8		0		0						10			250
9		0		0						0			250
10		0		0						0			250
11		0		0						0			250

* All blood sugars were performed at random by the John Malmros capillary blood sugar method, and expressed in milligrams per 100 cc of blood

R M male age 46 weight 257 A diabetic for 4 years he was originally converted to Orinase from 97 U of NPH insulin He continued sugar free on 1 0 gm Orinase daily for 7 months when his diabetes decompensated and he went back on insulin to 125 U daily His response to Diabinese was rapid and insulin was discontinued again Glycosuria recurred after 2 months and he was shifted to insulin and Orinase

CASE REPORT 3
Insulin Conversion

ORAL DIABETIC THERAPY – DIABINESE

Conver – Report

Patient Name: W W
Chart No: 3578

Date 1958	M	Urine Keto-Test A M	A M	P M	P M	NPH Insulin U-40 A M	A M	P M	P M	Dosage Per Day
May 30		BS*	216				20			200 mg
31							18			200
June 1							16			200
2							14			200
3							12			200
4							10			200
5							8			200
6							7			200
7							6			200
8							5			200
9							6			200
10							5			200
11		BS	158				0			200
	ued									

* All blood sugars were perfor- … l of n Malmros capillary blood sugar method and expressed in milligrams per 100 cc of blood

W W male age 68 weight 1 4 A diabetic for 14 years patient was taking 20 to 30 u of NPH insul n He was put on 20 units of NPH with 200 mg of Diabinese daily He continued sugar free gradually reducing his insul n and discontinu ng it in less than 2 weeks

CASE REPORT 4
Insulin Conversion and Withdrawal of Diabinese

ORAL DIABETIC THERAPY – DIABINESE
Conversion and Progress Report

Patient Name: A L
Chart No: 3667

Date	Urine Sugar Test				Urine Keto Test				Insulin				Dosage
1958	A M	A M	P M	P M	A M	A M	P M	P M	A M	A M	P M	P M	Per Day
May 3		4		2	BS*	144							500 mg
4		0		2									500
5		0		2									500
6		0		tr									500
7		0		0									250
8		0		0									250
9		0		0									250
10		0		0	BS	97							200
11		0		0									200
12		0		tr									200
13		0		tr									200
14		0		0									200
15		0		0									200
16		0		0									200
17		0		0									200
18		0		0									–
19		0		tr									100
20		0			BS	135							0
	Diabinese Discontinued												

* All blood sugars were performed at random by the Folin Malmros capillary blood sugar method, and expressed in milligrams per 100 cc of blood

CASE REPORT 5
Previously Untreated

ORAL DIABETIC THERAPY – DIABINESE

Conversion and Progress Report

Patient Name: H M

Chart No: 3668

Date	Urine Sugar Test				Urine Keto Test				Insulin				Dosage
1958	A M	A M	P M	P M	A M	A M	P M	P M	A M	A M	P M	P M	Per Day
May 22				0				tr	BS*	244			400 mg
23		1		1		tr		tr					400
24		1		1		0		0					400
25		1		1		0		0					400
26		1		0		0		0					200
27		0		0		0		0					200
28		0		0		0		0	BS	138			200
29		0		0		0		0					200
30		0		1		0		0					400
31		0		0		0		0					400
June 1		1		0		0		0					400
2		0		0		0		0					200
3		0		0		0		0					200
4		1		0		0		0					400
5		0		0					BS	202			400
6		0		0									200
7		0		0									200
8		0		0									0
9		0		0									
10		0		0									
11		0		0					BS	189			
Diabinese discontinued 6/8/58													

* All blood sugars were performed at random by the Folin Malmros capillary blood sugar method, and expressed in milligrams per 100 cc of blood

H M female age 47 weight 185 A diabetic for 2 years, and on diet without insulin Blood sugar 244 with 4 plus sugar She was put on Diabinese 200 to 400 mg daily, and tests became sugar free in a few days At the end of 1 week her blood sugar was 138, Diabinese was discontinued in 2½ weeks

EXPERIENCES WITH TOLBUTAMIDE AND CHLORPROPAMIDE IN TUBERCULOUS DIABETIC PATIENTS

Francis O Segarra, David S Sherman, and Benson S Charif
Boston Sanatorium Mattapan, Mass

Although the literature regarding the action, use, and results of treatment of diabetes with tolbutamide (Orinase) is voluminous and detailed, there has been very little recorded about the use of this drug in the treatment of diabetes coexistent with active pulmonary tuberculosis Inasmuch as these two diseases usually complicate each other, it seemed appropriate to study the effects of this oral antidiabetic drug in such cases When the discovery of chlorpropamide 1 propyl 3-(*p* chlorobenzenesulfonyl)urea was announced a comparative study of the two drugs seemed to be a logical and desirable project

Tuberculous Diabetic Patients Treatment with Tolbutamide, House Diet, and Antituberculosis Chemotherapy

A group of 9 patients with far advanced active pulmonary tuberculosis and coexistent diabetes mellitus were treated with tolbutamide and standard house diet (about 3000 calories per day) Although all patients in this group had active tuberculosis, none had any signs of toxicity such as fever, chills, emaciation or hemorrhage Tuberculosis treatment in all cases consisted of daily isoniazid (INH) and *p* aminosalicylic acid (PAS)

Seven patients were males, two were females their ages ranged from 42 to 83 years

Diabetic histories In three patients the diabetes had been present for several years One of these had been well controlled with 40 to 50 U of NPH insulin (case 1) The other two cases (2 and 4) had been controlled b diet without insulin

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fourth (case 7) required 75 U

In two patients the diabetes was discovered during hospitalization for tuberculosis One was found 21 days after admission (case 3) and the other, five months after admission (case 5)

diabetes, with only one requiring 75 U
nts had ever had an episode of acidosis
patients were not ideal candidates for
heir insulin requirements, they were
included in the study to determine if any benefit might be derived from oral antidiabetic therapy

Lab ere made before the start of therapy, after one week of the
A/G

(WBC) and fasting blood sugar (FBS) The sugar was checked by 4 urine sugar tests daily and by frequent FBS studies

The duration of tolbutamide therapy in all cases was too brief (22 days to 4 months) to determine any secondary failures Insulin was omitted the [illegible] 2 gm was given for 3 days [illegible] ere not satisfactorily con[illegible] increased to 4 gm daily

TABLE 1 represents a tabulated summary of this study The penultimate column indicates that only one patient (case 3) was satisfactorily controlled by tolbutamide In this very small series this represents a 12 per

TABLE 1
TUBERCULOUS DIABETICS FREE DIET AND TOLBUTAMIDE

	Patient	Sex	Age	Activity of tuberculosis	Diabetic history	Type of diabetes	Insulin units	Insulin control	Tolbutamide control	Duration on tolbutamide
1	B M	♂	63	Inact	4 yr	Mod	40 50	Good	Poor	22 days
2	B F	♂	75	Mod	9 yr	Mod	35 50	Good	Poor	3 mo
3	H C	♀	59	Act	1 mo	Mod	30	Good	Good	4 mo
4	J B	♂	83	Mod	1 yr	Mod	20–40	Good	Poor	3 mo
5	E M	♀	54	Act	2 mo	Mod	30–45	Good	Poor	3 mo
6	K N	♂	76	Mod	20 mo	Mod	40–50	Fair	Poor	1½ mo
7	J R	♂	57	Mod	9 mo	Mod	75	Good	Poor	3½ mo
8	J S	♂	42	Act	8 mo	Mod	35	Good	Poor	2 mo
9	C R	♂	70	Act	2 yr	Mod	55	Fair	Poor	2 mo

cent rate of success Therefore we conclude that diabetic patients who require a free high calorie diet for the treatment of their tuberculosis even if the diabetes is only of moderate degree are not good prospects for successful tolbutamide control In such cases insulin remains the drug of choice

In all the cases studied laboratory studies remained normal (with the exception of urine sugar and FBS in the failures) and no toxic side effects were observed

Tuberculous Diabetic Patients on Restricted 2000 Calorie Diet Treatment with Tolbutamide and Antituberculosis Drugs

Ten tuberculous diabetic patients were given a 2000 calorie diet containing fat 80 gm protein 100 gm and carbohydrate 200 gm (TABLE 2) Five of the patients in this group were also members of the first group (unrestricted diet) The same preliminary studies were made and the same tolbutamide dosage was given

Before starting tolbutamide the single-dose test was made in all ten patients

Except for case 7 who showed no response 4 hours following the ingestion of 3 gm of Orinase the results of the single-dose test varied from 18 to 52 per cent reduction of blood sugar with an average of 28 per cent

There was no correlation between the effect of the single-dose test and the

remaining six patients, who required more than 30 U of insulin, did not respond well

In the chlorpropamide group, 6 of the 9 patients had disease that was controlled with 30 U or less of insulin These six patients showed a good response to chlorpropamide, and were well controlled The remaining three cases had previously required more than 30 U for adequate control, and these patients did not do well on chlorpropamide

From these observations, we conclude that in our small groups of tuberculous diabetic patients the efficacy of both drugs is dependent on the mild or moderate nature of the disease Those patients requiring less than 30 U of insulin for control do well with either drug This conclusion is further supported by the fact that, of the seven patients who were studied in both drug groups the same patients did well with both drugs and the remaining patients did badly with both drugs

Although our studies were limited to a small number of patients, we feel that we can state as a clinical guide that a tuberculous diabetic patient, even with far advanced pulmonary disease who is not toxic and who is on a 2000-calorie diet will do well with either tolbutamide or chlorpropamide if his insulin requirement for control is 30 U or less On the other hand, patients requiring even a little more than 30 U failed to respond to either drug

The Action of Isoniazid on Carbohydrate Metabolism

Isoniazid (isonicotinic acid hydrazide) has been for the past several years one of the drugs commonly used in the treatment of tuberculosis There has been only rare mention of the effect of INH on carbohydrate metabolism and on the fasting blood sugar these reports have been primarily in the foreign literature We believe that our preliminary observations (TABLE 4) in this area might be of more than academic interest and that this subject merits further investigation

TABLE 4
HYPOGLYCEMIA EFFECT OF ISONIAZID

	Patient	Sex	FBS mg per cent	BS 4 hr after 500 mg INH		FBS mg per cent	BS 4 hr after tolbutamide 3 gm		FBS mg per cent	BS 4 hr after tolbutamide and INH	
				mg per cent	% decrease		mg per cent	% decrease		mg per cent	% decrease
1	J B	♂	327	285	13	394	293	24	259	171	34
2	A B	♂	191	182	5	217	130	40	222	126	43
3	K X	♂	327	285	13	318	259	19	370	309	17
4	C A	♀	450	443	0	490	490	0	467	467	0
5	H C	♀	201	146	26	176	130	26	163	101	39
6	E M	♀	234	154	34	271	154	43	278	118	57
7	A C	♀	420	359	15	443	349	21	348	285	18
8	J D	♂	182	201	10*	212	126	41	150	104	30

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In a group of 8 tuberculous diabetic patients, the following procedures were performed (1) tolbutamide single dose test using 3 gm of the drug, (2) a similar test using 500 mg of INH (with determination of the FBS before, and 4 hours after, ingestion of 500 mg of INH) and (3) a similar test using both 3 gm of tolbutamide *and* 500 mg INH given simultaneously

Results The tolbutamide single dose tests produced blood sugar reductions ranging from 19 to 43 per cent, with an average decrease of 28 per cent of the initial FBS level One patient showed no response

In the INH single-dose test one patient showed an increase in the FBS of 10 per cent The patient who showed no response to tolbutamide showed no response to INH The remaining 6 patients showed a significant reduction of the blood sugar, ranging from 5 to 34 per cent with an average reduction of 18 per cent

When the combined single-dose test (3 gm Orinase *and* 500 mg of INH) was given to the same group, there were reductions in the blood sugar ranging from 17 to 57 per cent with an average of 35 per cent

From these studies it is apparent that INH has a definite effect on carbohydrate metabolism in almost every case INH exerts a hypoglycemic effect that is not an insignificant one and when combined with tolbutamide, seems to enhance the action of this drug

These observations suggest an area of investigation that hitherto has not been reported in the search for oral control of diabetes

Summary

The efficacy of two oral antidiabetic agents in cases of diabetes with coexistent pulmonary tuberculosis (active but not toxic) was studied and compared These two agents are tolbutamide and chlorpropamide The salient observations resulting from this study are as follows

The results of treatment with tolbutamide and chlorpropamide in tuberculous diabetics are essentially the same

Tuberculous diabetic patients who require free diet ("house diet") because of their tuberculosis are not good candidates for treatment with either of the oral antidiabetic agents This is true even if the diabetes is mild

Tuberculous diabetic patients on a 2000-calorie diet show good response to both tolbutamide and chlorpropamide in cases of mild diabetes that were previously controlled by 30 U of insulin or less It is interesting to note that if insulin requirements were even slightly more than 30 U, neither drug was effective in maintaining adequate control

The extent of tuberculosis did not seem to influence the activity of either drug All patients studied have active disease but were not toxic

Preliminary studies of the effects of isoniazid, the most commonly used drug today in the chemotherapy of tuberculosis on the blood sugar, revealed a significant hypoglycemic effect in almost every case When compared with one of the oral antidiabetic drugs (tolbutamide), the hypoglycemic effect was greater than with either INH or the antidiabetic drug alone In other words isoniazid enhanced the hypoglycemic effect of the antidiabetic drug This suggests further possibilities for the oral drug control of diabetes

greatest fall in blood sugar levels occurred at the 60 min interval and continued to 120 min The mean percentage fall at 60 min was 30 per cent, with a range of 3 to 55 per cent At 20 min the mean percentage fall was 11 per cent, with a range from a 10 per cent rise to a 30 per cent fall At 40 min, the mean percentage fall was 23 per cent with a range from a 1 per cent rise to a 44 per cent fall It is to be noted that, at this time interval,

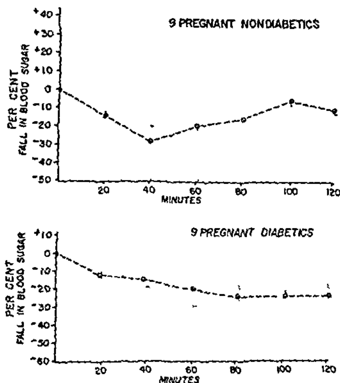


FIGURE 2 The mean tolbutamide tolerance curves of nine pregnant nondiabetics and nine pregnant diabetics Individual responses are recorded

the blood sugar levels of 3 patients (11 per cent of this group) were able to fall below the greatest mean percentage fall of the nondiabetic group Thus

of the diabetic group is characterized by a maximum at 60 min or more, and demonstrating no significant hypoglycemic responsiveness

The response to intravenous tolbutamide is not altered in pregnancy (FIGURE 2 upper graph) In a group of 9 nondiabetic pregnant patients, the greatest mean percentage fall was 28 per cent, and occurred at 40 min The range of variation was 4 to 52 per cent They also demonstrated a considerable degree of hypoglycemic responsiveness In the group of 9 preg

nant diabetic patients, the greatest mean percentage fall occurred at 80 min., with a range of 10 to 35 per cent Within 120 min there was no hypoglycemic responsiveness of any significance Of these 9 patients, only 1 whose glucose tolerance was abnormal demonstrated a normal tolbutamide response

CASE I

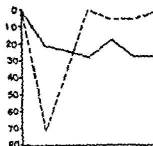
GLUCOSE TOLERANCE TESTS

	7 MONTHS	POSTPARTUM
F	86	113
1/2 HR.	160	105
1 HR.	172	135
2 HR.	142	112
3 HR.	130	102

TOLBUTAMIDE CURVE

——— INTRAPARTUM

- - - - POSTPARTUM



CASE II

GLUCOSE TOLERANCE TESTS

	7 MONTHS	POSTPARTUM
F	81	77
1/2 HR.	131	139
1 HR.	144	151
2 HR.	146	86
3 HR.	55	74

TOLBUTAMIDE CURVE

——— INTRAPARTUM

- - - - POSTPARTUM



FIGURE 3 Curves of two patients whose mildly abnormal glucose and tolbutamide tolerance at seven months' pregnancy was reversed completely in the postpartum period

In 2 instances, both glucose tolerance and tolbutamide curves were studied in the intra and postpartum periods (FIGURE 3) The upper part of the chart demonstrates a mildly abnormal glucose tolerance curve characterized by a blood sugar of 142 mg per cent at 2 hours The tolbutamide curve is also considered to be abnormal, demonstrating both a delayed fall and no significant return The postpartum glucose tolerance test shows a reversion toward normal The tolbutamide curve shows a completely normal response characterized by a rapid fall and a rapid return The lower section of the chart shows a similar situation where both abnormal glucose tolerance and tolbutamide curves at 7 months reverted to normal in the postpartum period.

Discussion

It is only in instances of suspicious mild diabetes, either in or out of p nancy, that it is difficult to arrive at a definite diagnosis Ur

cumstances additional measures beyond the glucose tolerance test have been wanting The response of the blood sugar in a 2 hour period to an intra venous *injection of sodium tolbutamide separates the mild diabetic from* the normal patient if 2 conditions are fulfilled first the fall in blood sugar should be prompt and should reach its lowest level by 40 min and second

promptly with a brisk fall in blood sugar levels the remainders of their curves are also in a downward direction In a very few instances was there a minimal irregular upward turn Analysis of the latter part of many more curves it is hoped will allow the degree of hypoglycemic responsiveness to be expressed mathematically We should like to emphasize the fact that Moyer s glucose tolerance criteria clearly separated the normal from the abnormal when correlating the tolerance curves with the tolbutamide curves There were no patients with blood sugars higher than 140 mg per cent at 2 hours who had a normal tolbutamide response Nine patients with dis tinctly abnormal tolbutamide curves had 2 hour blood sugars between 140 mg per cent and 160 mg per cent

The procedure is not associated with any significant side effects Only two patients had early in the test hypoglycemic symptoms that required the administration of glucose

We feel that further experience with this procedure will permit greater accuracy in detecting the mild diabetic state

Summary

The mild diabetic response demonstrates a lesser fall with no significant hypoglycemic responsiveness

(3) Pregnancy does not alter the tolbutamide response in normal patients

(4) It is felt that this procedure will help in detecting the mild diabetic state

Acknowledgment

We express our thanks to Martin G Goldner of the Jewish Chronic Disease Hospital Brooklyn N Y for his suggestions

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THE INTRAVENOUS TOLBUTAMIDE RESPONSE TEST IN THE DIAGNOSIS OF MILD DIABETES

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The striking difference in response to intravenously administered sodium tolbutamide exhibited by nondiabetics and mild diabetics has been reported in detail elsewhere [1,2] More recently the clinical usefulness of a standardized

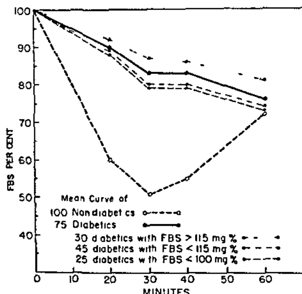


FIGURE 1 Mean tolbutamide response curves of all but five of the subjects studied

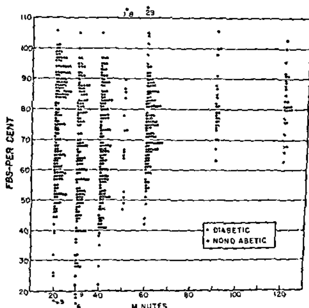
of the *Journal of Clinical Investigation* Volume 37 1958

intravenous tolbutamide response test in the diagnosis of mild diabetes mellitus has been delineated and diagnostic standards suggested [3] This paper therefore will merely review briefly certain aspects of these earlier reports as they pertain to the experiences of Zarowitz and Eis[4] with this test

Zarowitz and Eis have provided further valuable evidence to support the qualifications of the intravenous tolbutamide response test as a diagnostic procedure for mild diabetes In addition they have extended experience with the test to the pregnant state The correlation between reversible

abnormal tolbutamide response curves and reversible abnormal glucose tolerance curves in the intra- and postpartum periods, which they have observed, is consonant with the hypothesis that both tests measure the same physiological function

Clinical usefulness of a diagnostic test for mild diabetes mellitus depends on its ability *to differentiate the diabetic without fasting hyperglycemia from* the nondiabetic That the intravenous tolbutamide test qualifies in this respect is indicated by the data in FIGURE 1 It will be noted that the response curve of the mildest diabetics with fasting normoglycemia closely

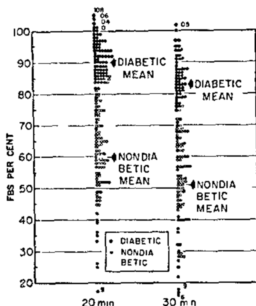


parallels the gently sloping curve of the more severe diabetics and is in distinct contrast to the rapid fall in blood glucose level typical of the nondiabetic group This fall reaches a nadir between 20 and 40 min and is generally followed by a vigorous rebound to or toward the pretest value Zarowitz and Eis report similar results

Zarowitz and Eis have emphasized the diagnostic value of the rebounding ... which, as they point out, *seldom occurs except* ... test beyond the 40 min point ... ve While there is no doubt as ... unds in the nondiabetic group, we have regarded this phenomenon as being secondary to the more rapid decline in blood glucose level exhibited by the group, that is, the result of counterregulatory forces thus set into play Rebound, in our opinion

reflects both the rate and extent of decline in blood glucose level and the ... rather than beta cell func

be expected to mirror most directly beta cell responsiveness to this stimulus



In FIGURE 2 in which the tolbutamide response curves of 100 nondiabetics and 75 mild diabetics are recorded maximum separation of the 2 groups occurs at 20 and 30 min A close up of these points is provided in FIGURE 3 At 20 min 95 per cent of the diabetic group have a blood glucose level of 80 per cent or more of the pretest value whereas 96 per cent of the nondiabetic group have a level of less than 84 per cent of that value Selection of 84 per cent and more as the diabetic zone and 80 per cent and less as the nondiabetic zone has provided standards that have thus far been 95 per cent accurate The zone between 80 and 84 per cent of the pretest level is considered presumptively abnormal but nondiagnostic *

* These standards apply only if blood glucose is determined by the Somogyi Nelson technique It is hoped that the data of Zarowitz and Eis will provide diagnostic standards for the Folin Wu method

The 30-min specimen (FIGURE 3) appears to be diagnostically more specific inasmuch as 99 per cent of the nondiabetics are at less than 77 per cent of the pretest level by this time However, this specimen is far too insensitive to be of value in the exclusion of diabetes, since 15 per cent of mild diabetics have reached the nondiabetic zone at this point

It would seem that the blood glucose response within the first 30 min of the tolbutamide injection provides the necessary diagnostic information in the vast majority of individuals Termination of the test at this point

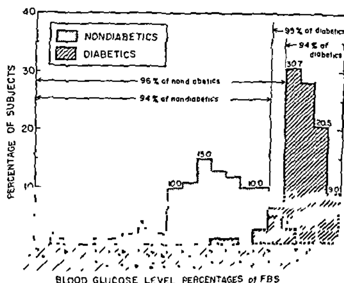


FIGURE 4 The frequency distribution of the blood glucose levels [in percentages of ... 20 min after ... per cent range ... with the per

endows it with an asset unique among diagnostic tests for mild diabetes, namely, brevity Furthermore, it shortens the period of hypoglycemia that occurs in the nondiabetic group If, however, the data of Zarowitz and Eis should indicate that examination of the rebound phase of the curve provides an unmistakable increase in diagnostic accuracy, prolongation of the test may be necessary

As in the case of the glucose tolerance test[5,6] it would be naive to expect that the tolbutamide response test can provide a sharp demarcation between

zone of overlap includes 10 per cent of each group and extends from 75 to 89 per cent of pretest level Within this zone rigid adherence to the recommended diagnostic criteria can lead to error since only probability of disease can be estimated For example these limited data suggest that among persons whose levels fall to between 75 and 79 per cent of the pretest value the diagnostic probability of diabetes is 21 per cent between 80 and 84 per cent of pretest level it is 56 per cent and between 85 and 89 per cent it is 91 per cent Above and below the overlap zone diagnostic accuracy approaches 100 per cent

It is our tentative impression that the zone of overlap of the tolbutamide response test is somewhat smaller than that of the oral glucose tolerance test due primarily to fewer false positive results It would be premature however to pass final judgment on the standards proposed for the tolbutamide response test in view of the controversy that still prevails in respect to the oral glucose tolerance test despite several decades of extensive use

At present however the intravenous tolbutamide response test appears to qualify as an accurate safe rapid and not unpleasant diagnostic procedure for mild diabetes that compares favorably with other methods now commonly used for this purpose

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A CLINICAL COMPARISON OF CHLORPROPAMIDE AND TOLBUTAMIDE*

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Chlorpropamide, a recent orally administered hypoglycemic agent, has been compared to tolbutamide with regard to acute and chronic hypoglycemic potency and clinical effectiveness The chemistry and animal studies of these two sulfonylureas are discussed elsewhere in this monograph

Material

Studies were conducted on 26 subjects including 1 normal and 25 diabetic Five of the diabetics were hospitalized and maintained throughout their course on a constant diet of known composition with a daily intake of carbohydrate ranging from 150 to 220 gm and protein from 80 to 120 gm Both *acute and chronic potency of the drugs were studied under these conditions The normal subject was allowed a free diet between comparative studies of* the 3 0 gm dose Sufficient time was allowed between the acute dosages of each drug to prevent interference with the subsequent hypoglycemic response *One of the diabetics had a previous total bilateral adrenalectomy* and another had diabetes secondary to a total pancreatectomy Sixteen of the diabetics were studied for 5 months as outpatients in the diabetes clinic of the University of California School of Medicine

Methods

Blood sugars were measured by the method of Nelson and Somogyi[1] Urinary glucose was measured by the glucose oxidase method[2] Serum chlorpropamide levels were determined by a procedure developed by the Chemical Research and Development Department of Chas Pfizer & Co, Inc Brooklyn, N Y[3] *The content of unesterified fatty acids in the blood* was obtained by the Davis extraction procedure,[4] followed by titration according to the method of Dole[5]

Results

Acute hypoglycemic effects Various amounts of chlorpropamide and tolbutamide were administered as a single dose to both normal and diabetic subjects in order to compare the acute hypoglycemic effectiveness of each Serum values of chlorpropamide and blood sugar levels were measured

* The study *reported in this paper was aided by a grant from* Chas Pfizer & Co, Inc, Brooklyn N Y

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simultaneously to study the acute relationship between the hypoglycemic effect and the concentration of the drug in the blood and to observe the degree and rapidity of absorption In FIGURE 1 the effect of the 0 5 gm dose on 3 diabetic patients is noted The blood sugars are measured in the fasting state for the first 4 hours It is apparent that the onset of hypoglycemia is within the first 2 hours with both drugs and continues for 4 hours

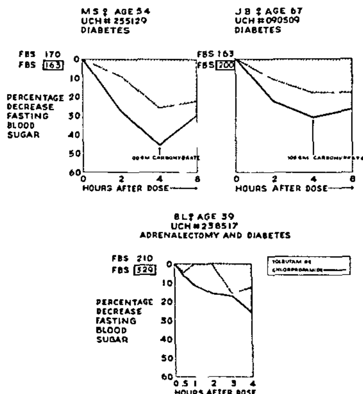


FIGURE 1 Comparative acute hypoglycemic potency of chlorpropamide and tolbutamide following 0 5 gm as single oral dose

before the standard meal is given Postprandially there is a rise toward base line level The comparative hypoglycemic potency of chlorpropamide is greater in each case than that of tolbutamide by a factor approaching 2 when the areas representing blood sugar fall are compared In the lower part of FIGURE 1 the effect of a similar dose given orally to an adrenalectomized patient maintained on cortisone replacement alone shows a minimal response to tolbutamide becoming significant 3 hours after the dose is given

1 0-GM P.O. SINGLE DOSE

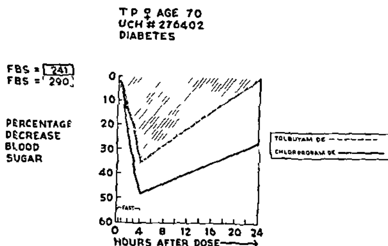


FIGURE 2 Comparative acute hypoglycemic potency of chlorpropamide and tolbutamide following 1 0 gm as single oral dose

3 0-GM PO SINGLE DOSE

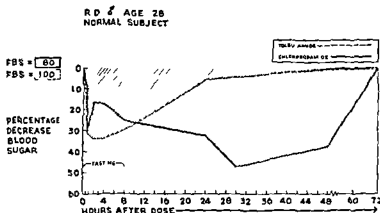


FIGURE 3 Comparative acute hypoglycemic potency of chlorpropamide and tolbutamide following 3 0 gm as single oral dose

Chlorpropamide, however, causes a much more significant fall starting within 30 min after oral administration Here the acute hypoglycemic potency of chlorpropamide is greater than that of tolbutamide by a factor greater than 2

FIGURE 2 illustrates the comparative effect of the 1 0-gm dose of the

2 compounds Again it may be noted that chlorpropamide has a hypo glycemic potency approaching 2 times that of tolbutamide when the areas of hypoglycemia for similar time intervals are measured

In FIGURE 3 the 3 gm dose of each compound administered to a normal subject demonstrates that at this dose level the comparative hypoglycemic potency of chlorpropamide is greater than 2 times that of tolbutamide when

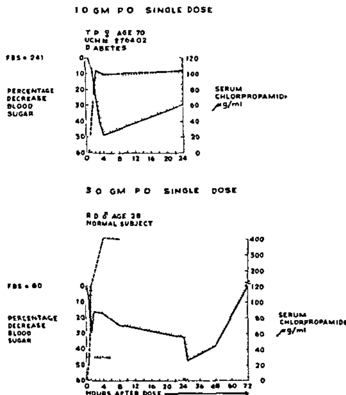


FIGURE 4 Serum levels of chlorpropamide compared to changes in blood sugar at the 1 0 gm dose (upper) and 3 0 gm dose (lower)

the total areas of blood sugar fall below the base line are compared How

in blood sugar below base line levels that lasts less than 48 hours

It would appear that the intrinsic potency of each drug has reached a maximum at this dose level and that their hypoglycemic effects are equal, acutely Their over all hypoglycemic potency, however, varies with the differences in duration of activity

Blood levels of chlorpropamide and its hypoglycemic effectiveness The serum levels of chlorpropamide measured simultaneously with blood sugar levels (shown in FIGURE 4) follow the 1 0 gm and 3 0 gm single oral dose It may be noted that the rate of increase and levels achieved for the serum values of chlorpropamide are proportional to the dosage These suggest that absorption is rapid

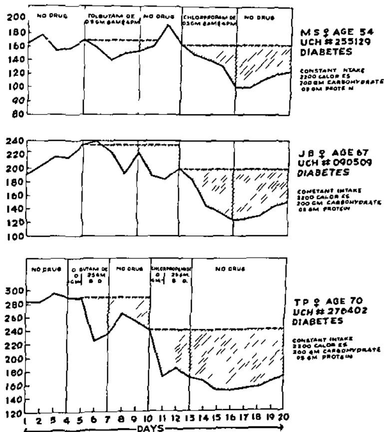


FIGURE 5 Comparative chronic hypoglycemic potency of chlorpropamide and tolbutamide compared at similar dose levels and time intervals

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... milarly obtained plasma values for tolbutamide in 2 previous studies

demonstrated by the fact that, as compared to tolbutamide, a lower serum value is required to achieve a significant hypoglycemic effect

Chronic hypoglycemic effects The comparative hypoglycemic potency upon repeated daily administration was studied in 3 hospitalized diabetic patients maintained on the metabolic ward on constant dietary intake for prolonged periods FIGURE 5 demonstrates that the relative potency of chlorpropamide is again greater than that of tolbutamide in all of the individuals studied Upon interruption of intake of each drug the hypoglycemic effect of the former persists longer, and fasting blood sugars remain below their original levels for 4 days In patient T P 7 days after stopping the drug the fasting blood sugar levels had not returned to their previous base line levels Twenty four hour urine sugars measured in these studies were generally below 100 mg for 24 hours and were not changed significantly during therapy The comparative areas of hypoglycemia reveal that the chronic potency of chlorpropamide upon repeated daily administration is of the order of 6 times that of tolbutamide

Rate of disappearance of chlorpropamide The apparent discrepancy between acute and chronic potency of chlorpropamide is most probably related to the prolonged rate of disappearance of this compound in the blood Half life periods were calculated based on the serum level of chlorpropamide following cessation of medication TABLE 1 shows that the half life rate of disappearance of chlorpropamide in 3 patients at different dose levels ranges from 36 to 39 hours In 2 previous studies tolbutamide revealed a half life ranging from 4 to 6 hours as calculated from the disappearance of the compound from the plasma [6, 7] These figures are close to the sixfold increase in chronic potency for chlorpropamide compared to tolbutamide previously shown In TABLE 1 we note that the serum level of chlorpropamide following repeated daily administration at doses of 0 5 gm daily and 200 mg 3 times daily remains steady during continued administration It is apparent that the serum levels of chlorpropamide have risen to more than the effective levels previously determined to be between 20 and 120 μg ml In addition the levels reach a plateau with repeated daily administration and remain relatively constant thereafter Thus after 72 and 107 days of treatment respectively in the two cases serum levels remain between 250 and 350 μg ml throughout Fasting blood sugar levels are not related to serum levels of chlorpropamide once the steady levels have been reached although good control of hyperglycemia has been achieved in each case

Mechanisms of action The possible mechanisms of action of this new sulfonylurea compound appear to be similar to those of tolbutamide and the other sulfonylureas previously studied Thus, in FIGURE 6 we note that the fasting blood sugar levels are not significantly affected in a depancreatized diabetic who received first saline and then chlorpropamide intravenously This is in agreement with previous studies on other sulfonylurea compounds in animals[8] and in man [9] Repeated daily administrations of chlorpropamide

TABLE 1

RELATIONSHIP OF ORAL DOSAGE TO SERUM LEVELS OF CHLORPROPAMIDE AND TO BLOOD SUGAR LEVELS

Case	Subject	Dosage	Duration of therapy	Time after stopping dose hours	Serum levels of chlorpropamide μg./ml.	Calculated rate of disappearance of chlorpropamide (half life)	Fasting blood sugar
Drug elimination							
1	B. K. Normal	1.0 gm.	single dose	0	0		80
				0.5	0		87
				1	31		55
				2	91		67
				4	114	38 hours	104
				6	164		96
				8	137		68
				24	102		82
2	R. D. Normal	3.0 gm.	single dose	0	0		80
				4	400		68
				8	400		57
				24	420	39 hours	54
				32	205		43
				48	180		50
				72	112		92
3	J. B. Diabetes	0.5 gm. twice daily	start day 1		0		200
			start day 2		67		182
			start day 3		116		155
			start day 4		187	36 hours	135
				16	177		125
				60	69		130
				84	48		150
Chronic drug levels							
4	L. S. Diabetes	500 mg. daily	start day 1		0		200
			start day 11		350		138
			start day 23		250		88
			start day 45		320		118
			start day 74		260		130
			start day 107		252		99
5	P. P. Diabetes	200 mg. thrice daily	start day 1		0		242
			start day 4		280		133
			start day 32		320		94
			start day 51		250		81
			start day 72		310		125

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administration ... do appear to ...
are not a direct effect of the sulfonylurea

... to that of
glycemic
diabetic

agent alone ...
acidosis, with wide variations in daily levels of fasting blood sugars and in

their daily dosage of insulin All 3 were found to be primary failures on chlorpropamide therapy at doses ranging from 0 5 gm to 1 5 gm daily and demonstrated steadily increasing blood sugar levels and ketosis within the first 3 to 4 days of therapy It is apparent therefore that functional beta cells are required for the action of chlorpropamide just as they are for the other sulfonylureas

Finally, chlorpropamide was given in repeated daily doses to an adrenalectomized patient maintained on cortisone alone In FIGURE 7 a rather exaggerated hypoglycemic response may be observed The 24 hour urinary

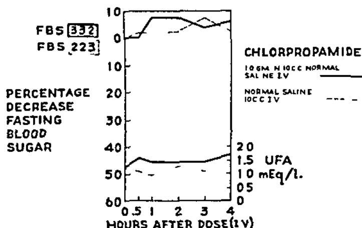


FIGURE 6 Acute effects of chlorpropamide (and a similar volume of saline given intravenously) on the fasting blood sugar and unesterified fatty acid levels of a depancreatized diabetic

sugars shown in the lower part of the figure parallel the fasting blood sugars in onset and duration of fall This suggests that chlorpropamide may have a depressant action on the gluconeogenesis induced by steroid administration an important factor in this patient's hyperglycemia

Outpatient trials A limited number of stable diabetics was given chlorpropamide in the diabetes clinic Criteria for selection of patients were the same as those used to select candidates for tolbutamide therapy None had

had been treated on dietary regimen alone without success Three patients had been well controlled on small doses of insulin and 8 patients had been on tolbutamide previously with good control and no untoward effects It

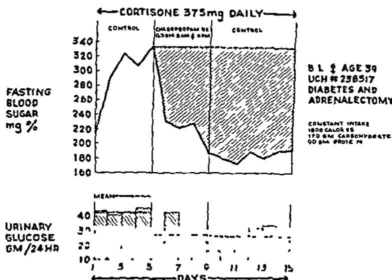


FIGURE 7 Increased sensitivity and prolonged hypoglycemic effect of chlorpropamide administration to an adrenalectomized diabetic patient

was found that 7 patients complained of nausea soon after starting chlorpropamide Doses in excess of 600 mg daily were consistently associated with nausea, reducing the dosage below this level was generally followed by relief of symptoms Four patients however, had to be returned to their [illegible]

TABLE 2
OUTPATIENT TRIALS ON CHLORPROPAMIDE

Total number of patients	16	
Unstable diabetics (all primary failures)	3	
Stable diabetics		13
Previous dietary treatment alone	2	
Previous insulin	3	
	8	
		7
		4
		9
	2	
	2	
Previous tolbutamide	5	
Dosage tolbutamide	1 to 2 gm daily	
Control	good	
Untoward symptoms	none	
Duration of treatment 6 months to 2 years		
Dosage, chlorpropamide	250 to 600 mg daily	
Control	good	
Untoward symptoms	none	
Duration of treatment 3 months to 6 months		

from 3 to 6 months of treatment Repeated hematologic, hepatic, and renal studies have revealed no toxic effects of the drug to date

In our clinical experience the maintenance dose of chlorpropamide was found to be one third to one quarter that of tolbutamide However con sidering the chronic potency and the difference in the rate of disappearance from the blood a relative dosage ratio of 1 5 or 1 6 should prove effective further clinical studies may show this to be so

Discussion

The present study suggests that chlorpropamide is a significantly more potent hypoglycemic agent than tolbutamide both in acute and chronic activity The former appears to be determined merely by the intrinsic

half life of the drug in body fluids in its active form This was found to be 36 hours instead of the 6 hours for tolbutamide, and might thus account for the difference in chronic potency between the 2 It was interesting to note that the serum level of chlorpropamide reached a relatively steady plateau not exceeding 350 µg per ml following prolonged periods of administration at the dosage of 0 5 gm daily

As to the mechanism of action, chlorpropamide would appear to require the presence of functioning beta cells of the pancreas just as do the other sulfonyl ureas previously studied Chlorpropamide did not effect a decrease in blood

ones showing that higher dosages of sulfonylureas than those necessary to produce hypoglycemia may inhibit insulin degradation [12]

A slightly exaggerated hypoglycemic effect observed in an adrenalectomized patient on cortisone therapy suggests that chlorpropamide may al o be an inhibitor of action on gluconeogenesis

The maintenance dosage of chlorpropamide appears to lie between one third and one sixth that of tolbutamide From our experience we conclude that if this is kept at less than 500 mg per day, the incidence of nausea is markedly reduced

Summary

Chlorpropamide has approximately twice the hypoglycemic activity of tolbutamide in its acute effect for a four hour period

ac

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to be six times greater than that of tolbutamide

The mode of action of the two sulfonylurea derivatives appears to be identical

The prolonged action and tendency to nausea, when 500 mg daily is exceeded suggest that care must be observed in its clinical use

Acknowledgments

We acknowledge the cooperation of Herbert C Moffitt, Jr, *chief of the* diabetes clinic of the University of California Department of Medicine Robert Reiss and other members of the diabetes clinic We are indebted to Richard Havel for the studies mentioned under *Methods*

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EXPERIENCES WITH CHLORPROPAMIDE WITH REFERENCE TO ITS INTERMITTENT USE

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Department of Medicine American University of Beirut Lebanon

Our experiences with chlorpropamide were obtained in the Middle East where there are two reasons for a tremendous need for an oral antidiabetic preparation First the standards of hygiene and the habits of living make the use of insulin difficult if not hazardous Second the people are accustomed to a gigantic consumption of carbohydrate Arabic bread is indeed the staff of life and carbohydrate is consumed at the rate of 500 to 800 gm /day If the production of other foodstuffs were feasible from an economic and cultural point of view many diabetics might be controlled by weight reduction and a shift in the source of calories This is not now possible hence an oral antidiabetic agent is most valuable

TABLE I
BLOOD LEVELS OF CHLORPROPAMIDE* IN MG PER CENT ON A DAILY ORAL INTAKE OF 1 GM AND CESSATION ON 8TH DAY

Patient No	Days											
	1	2	3	4	5	6	7	8	9	10	11	12
13	5 8	5 9	12 2	9 2	22	26	18	15 6	9	10	5	4
14					20	23	15	10	4 5	5 5	2 5	1 5
17	6 3	11	12	14	16	14	13	11	12	6 8	—	2 5

* Supplied by Max Gahwyler of Chas Pfizer & Co Inc Brooklyn N Y

Chlorpropamide has certain characteristics that apart from its hypoglycemic qualities make it of special use in the management of diabetes Upon the daily oral administration of a fixed amount the blood level rises gradually to a maximum in 5 to 7 days If the administration is then interrupted the blood level falls very slowly so that there may be a concentration of 4 to 5 mg per cent even after 5 days This is illustrated in the figures furnished by Gahwyler on blood levels (TABLE 1) This suggested the possibility that the drug could be given intermittently

Our patients were admitted to the hospital where diet and activity could be controlled The patients selected the diet they thought they could maintain for the period of st [illegible]

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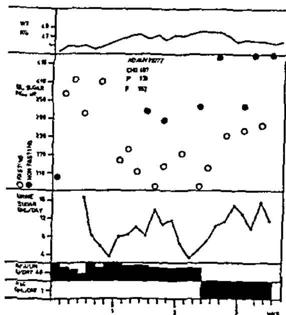


FIGURE 1 Unstable juvenile diabetic.

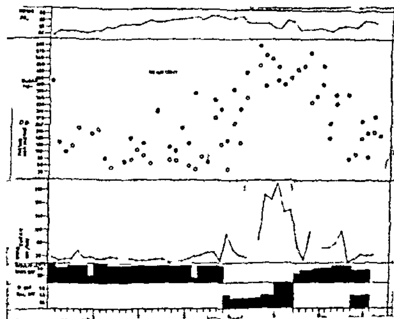


FIGURE 2 Severe diabetic with renal glycosuria

Our detailed studies with chlorpropamide are illustrated by the following cases

The first patient was an 18 year old girl (N D 99277) with diabetes of 7 months duration chlorpropamide did not control her diabetes in any way (FIGURE 1)

The second patient was a 50 year old farmer (M S 126347) with diabetes of 4 years duration and a low renal threshold for sugar His diabetes was severe and again could not be controlled by chlorpropamide (FIGURE 2)

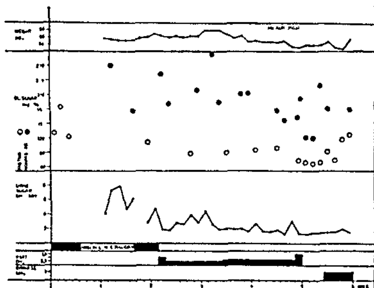


FIGURE 3 Mild diabetic

It was interesting to note that there was no apparent effect on his renal glycosuria The concomitant use of chlorpropamide with insulin did not appear to reduce the insulin dosage substantially

The third patient was a 60 year old a former wrestler (M K 125037) with mild diabetes for four years (FIGURE 3) He had been taking 20 units of insulin irregularly He had proteinuria slight edema and a slightly low serum albumin of 2 9 gm per cent After a period of stabilization on insulin he was changed to chlorpropamide On a daily maintenance dose there was no evidence of hypoglycemia but the interesting feature was that a single dose of 1 5 gm maintained his blood sugar and 24 hour urine excretion within normal limits for 72 hours and without hypoglycemia (FIGURE 4) There was no evidence of acceleration of the diabetic vascular damage while on chlorpropamide His evegrounds proteinuria, and hypoalbuminemia remained constant His urea nitrogen fluctuated between 15 and 27 mg per cent

The fourth patient was a 50 year old man (K D 89351) with diabetes of

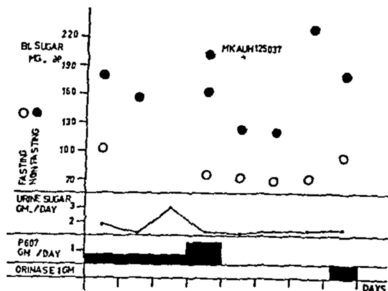


FIGURE 4 Mild diabetic with prolonged response to chlorpropamide

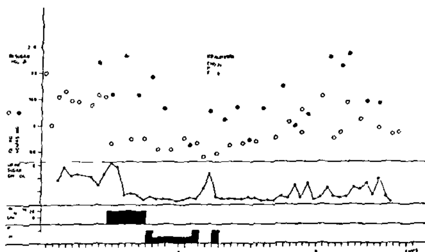


FIGURE 5 Mild diabetic with prolonged response to chlorpropamide

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very small doses of chlorpropamide given ... trolled his blood and urine sugar

The fifth patient was a 60 year-old woman with diabetes of four months

duration Difficulties in her control by intermittent chlorpropamide are shown in FIGURE 6

The studies have been extended to include our clinic patients whose diet and activity cannot be controlled In five patients taking 0.5 gm of chlorpropamide every two to three days the nonfasting blood sugar could frequently be maintained at the upper limits of normal However, this was much less striking than in the third patient (M K 125037), whose 72 hour control probably represented the delayed excretion of a cumulative dose On

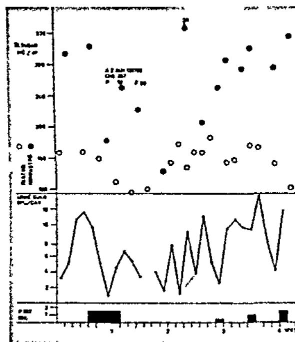


FIGURE 6 Mild diabetic with inadequate response to intermittent chlorpropamide

the other hand, our fourth patient (K. D 89351) maintained normal values for a very long time after a relatively small dose of chlorpropamide The fifth patient represents some of the difficulties inherent in attempting intermittent dosage

We have not had the opportunity to study the specific effects of this drug on the complications of diabetes In the cases studied, there was no evidence for any acceleration of the degenerative processes in retinal, coronary, or renal circulations

The intriguing aspect of chlorpropamide is its mechanism of action particularly, how this drug can, while circulating at slowly decreasing blood levels maintain the blood sugar within normal limits despite three ingestions of carbohydrate a day

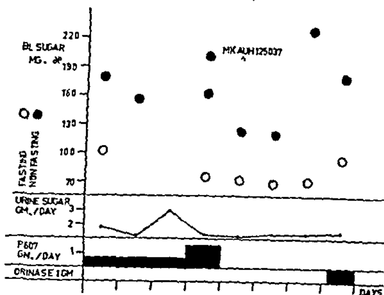


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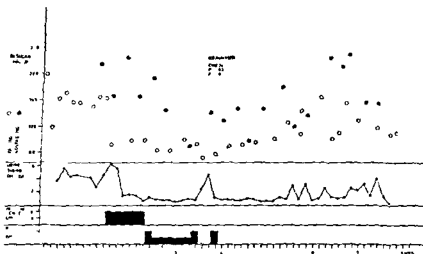


FIGURE 5 Mild diabetic with prolonged response to chlorpropamide

2 years' duration He was a mild diabetic and of a comfortable, middle-aged habitude that resists all changes in weight (FIGURE 5) On his particular diet the need for 20 units of insulin is demonstrated, and it is apparent that very small doses of chlorpropamide given at considerable intervals controlled his blood and urine sugar

The fifth patient was a 60-year-old woman with diabetes of four months'

duration Difficulties in her control by intermittent chlorpropamide are shown in FIGURE 6

The studies have been extended to include our clinic patients whose diet and activity cannot be controlled In five patients taking 0 5 gm of chlorpropamide every two to three days the nonfasting blood sugar could frequently be maintained at the upper limits of normal However this was much less striking than in the third patient (M K 125037) whose 72 hour control probably represented the delayed excretion of a cumulative dose On

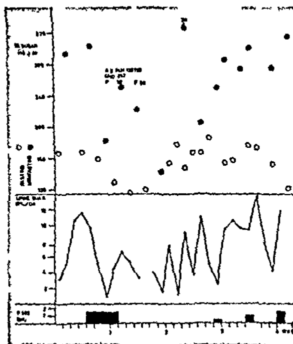


FIGURE 6 Mild diabetic with inadequate response to intermittent chlorpropamide

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The intriguing aspect of chlorpropamide is its mechanism of action particularly how this drug can while circulating at slowly decreasing blood level maintain the blood sugar within normal limits despite three ingestions of carbohydrate a day

Conclusion

In occasional patients the slow excretion of chlorpropamide may permit its intermittent use

Discussion of the Paper

J D N NABARRO (*Middlesex Hospital London England*) From the purely clinical standpoint the question that must be answered is Does chlorpropamide have any advantages over tolbutamide? Chlorpropamide is certainly easier for the patient to take and it can be given once daily whereas tolbutamide must be given in divided doses

A major problem with tolbutamide at the present time is the development of acquired or secondary resistance to its hypoglycemic action This is not the same as secondary failure a term used by several contributors to this monograph Secondary failure is more often due to the fact that the patient has broken his diet than to acquired resistance to the hypoglycemic drug and it is not reasonable to blame the drug for the indiscretions of the patient Acquired resistance can be established only if the patient is hospitalized and remains hyperglycemic while on a strict diet or if a high dosage of tolbutamide (3 gm /day) can be withdrawn without any further rise of blood sugar Our incidence of acquired resistance is 10 per cent for patients on the drug for more than 6 months and it has developed between the seventh and seventeenth months of treatment One patient with acquired resistance was transferred to chlorpropamide without adequate response

We have had to withdraw tolbutamide in 7 per cent of our patients because of side effects 4 per cent abdominal and 3 per cent skin changes One patient who was unable to take 1 5 gm of tolbutamide because of abdominal discomfort is now well controlled without symptoms on 0 25 gm of chlorpropamide daily

One final point that I should like to make is that we must not underestimate the length of time required for proper assessment of the value of these drugs We have recently examined all our data on tolbutamide treated patients and have found that the percentage still on the drug and in satisfactory control drops steadily over the first 18 months However between 18 months and 3 years the figure levels out at about 35 per cent of those giving a good initial response This of course tells us nothing about the antidiabetic value of the drug and it relates only to its hypoglycemic action

A CLINICAL AND PHARMACOLOGICAL COMPARISON OF CHLORPROPAMIDE AND OTHER SULFONYLUREAS*

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The effective blood level of chlorpropamide† probably lies between 4 and 16 mg per 100 ml, which is comparable with those of carbutamide and tolbutamide, but chlorpropamide differs significantly from tolbutamide in that it is not readily metabolized to a physiologically inactive compound as tolbutamide is to carboxytolbutamide.[1] In fact, the ultraviolet absorption spectrum of chlorpropamide in the blood seems to be identical with that of the drug in the urine, suggesting that it has undergone no chemical change.

TABLE 1
MEAN HALF LIFE AND PERCENTAGE OF BODY WEIGHT IN WHICH THREE SULFONYLUREAS WERE DISTRIBUTED

	Half life hours	Volume of distribution as percentage of body weight
Carbutamide	40 (8)	approx. 45
Tolbutamide	3.5 (6)	approx. 20
Chlorpropamide	34.5 (8)	approx. 18

Numbers of individuals in parentheses.

This effective blood level of chlorpropamide can be maintained with as little as 0.5 gm/day in contrast to the three- or fourfold dosage that is required for tolbutamide. Both drugs are rapidly absorbed; they produce peak serum levels 2 to 3 hours after ingestion. The volume of distribution of both drugs in the body is similar (TABLE 1) and approximates the volume of the extracellular fluid, whereas carbutamide is distributed in about twice as large a volume. The half life of chlorpropamide, that is, the time required for any given serum level to fall by 50 per cent, is approximately 10 times longer than that of tolbutamide and nearly as long as that of carbutamide (TABLE 1). FIGURE 1 illustrates how these data were obtained. A single dose, for example 2 gm, of a sulfonylurea is given to an individual who has not previously received this drug. Blood samples are then withdrawn at intervals determined by the anticipated half life, and the concentrations of the drug are estimated in these samples using a controlled serum taken before the experi-

ment started When these concentrations are plotted on a *log scale against* time a straight line is obtained This line can then be extrapolated back to zero time, and the theoretical initial concentration is then read from the *scale* The assumption is made that all the drug administered was absorbed and that no significant fraction was excreted before absorption was complete *Having thus derived the theoretical initial concentration* the time at which

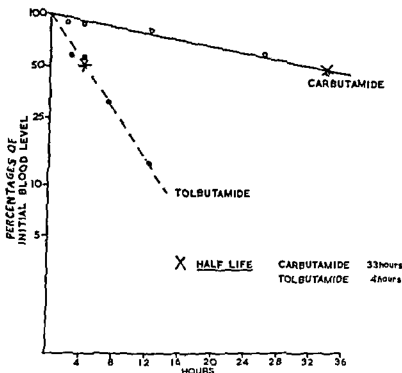


FIGURE 1 Rate of fall in serum level of carbutamide and tolbutamide after a single administration of 3 gm of each to the same individual at different times The theoretical initial concentration is obtained by extrapolating the line representing the rate of decrease of blood levels recorded on a log percentage scale Reproduced by permiss on of *The Lancet*

[illegible] ll have fallen by 50 per cent can be read from the graph In the example

[illegible]

The volume of distribution of the drug can readily [illegible] the theoretical initial concentration *and the size of the dose administered* [illegible] absorption was complete [illegible] uted in the body in a volume [illegible] d it is of interest to know whether it is freely in solution or bound to protein There is some evidence that it is protein bound because serum containing *15 mg/100 ml of chlor*

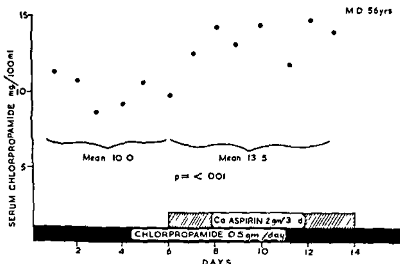


FIGURE 2 Effect of salicylate on chlorpropamide serum level

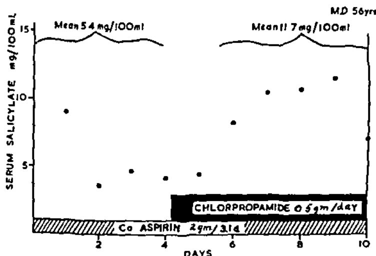


FIGURE 3 Effect of chlorpropamide on serum salicylate level

propamide had all traces of chlorpropamide removed by the precipitation of the protein with trichloracetic acid and neither chlorpropamide nor tolbutamide has been found in the cerebrospinal fluid when the serum concentration has been as high as 14 to 24 mg /100 ml The protein binding is evidently a loo e binding because chlorpropamide can be dialyzed out of serum quite readily The difference in the half lives of tolbutamide and

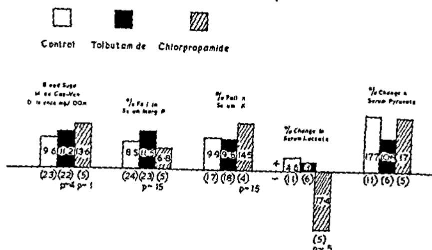


Figure 4 Changes during glucose tolerance tests (50 gm *per os*) in mild diabetics Numbers are shown in brackets

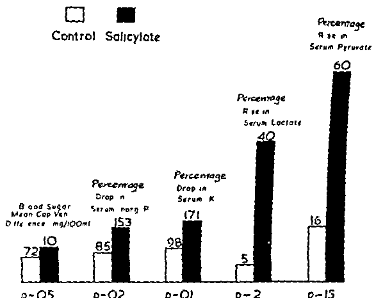


Figure 5 Changes during glucose tolerance tests (50 gm *per os*) in 11 mild diabetics

chlorpropamide must depend largely on their handling by the kidneys, since the urine is the vehicle of their elimination The protein binding itself would tend to limit the renal excretion of those drugs, but the mechanism of tubular transport must be the key to the understanding of this problem Some interesting sidelights were thrown on it by studying the interaction of chlorpropamide and salicylates[2] on blood levels, first when given singly and

then when given together to the same individual (FIGURES 2 and 3). Such results are explained most easily by mutual interference with some process of tubular secretion in the kidney, and may have some practical bearing on therapy, since their hypoglycemic effects can be shown to be additive.

In comparing the hypoglycemic potency of tolbutamide and chlorpropamide, the superiority of chlorpropamide is apparent notwithstanding the small number of cases that we have properly studied (TABLE 2). At blood levels that were slightly lower, chlorpropamide produced a 61 per cent fall in the fasting venous blood sugar compared with a 29 per cent fall induced by tolbutamide. Indeed, so great is the hypoglycemic potency of chlorpropamide that we have twice been troubled by hypoglycemia induced in mild diabetics, a feature we have rarely experienced with tolbutamide. Unlike

TABLE 2
HYPOGLYCEMIC EFFECT OF TOLBUTAMIDE AND CHLORPROPAMIDE
Mild diabetics (numbers in parentheses)

	Mean percentage fall in fasting blood sugar		Mean drug level mg/100 ml
	Drug day	No treatment day	
Tolbutamide	29	(25)	12·4
Chlorpropamide	61	(5)	11·4
	$p < 0.005$		

our experience with tolbutamide, we have never found chlorpropamide to be even probably responsible for dyspepsia. We have had three patients who had had dyspepsia, one an actual case of hematemesis that we attributed to tolbutamide. All three were changed over to chlorpropamide and made no further complaints about their digestion. The less frequent administration of chlorpropamide in itself is no doubt helpful in this respect.

Data have been obtained on patients taking tolbutamide and chlorpropamide during oral glucose tolerance tests and compared with similar data obtained when they received no treatment. In this way it was hoped that some information might be gained regarding the method of the hypoglycemic action shown. The parameters measured were the differences between the capillary and venous blood sugar levels,[3] the fall in the serum inorganic phosphate[4] and potassium,* and the change in the serum level of lactate[5] and pyruvate,[6] all estimated six times during each glucose tolerance test. The results are expressed in FIGURE 4.

If there had been an insulinlike action in the peripheral tissues characteristic changes would have been found in each of these different measurements. In fact, no significant changes were observed and thus no evidence was obtained that these sulfonylureas acted like insulin in the muscles. The fact

* Internal standard flame photometry.

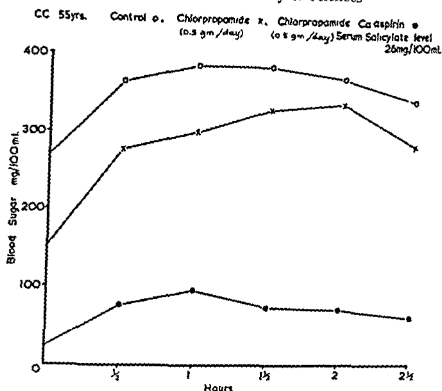


FIGURE 6 Glucose tolerance curve obtained in the same patient when on no treatment, when receiving chlorpropamide (one half gm /day), and when receiving the same dose of chlorpropamide with calcium aspirin sufficient to bring the serum salicylate level to 26 mg 100 ml

that relatively they have much more effect on the fasting than on the post-absorptive blood sugar levels suggests that they inhibit glucose output by the liver
(FIGURE 5)
significant,

TABLE 3
BLOOD GLUCOSE LEVELS IN 4 MILD DIABETICS
Glucose Tolerance Tests 50 gm *per os*

	Fasting venous blood glucose mg /100 ml				Peak venous blood glucose mg /100 ml			
Patient	M D	M K	H C	C C	M D	M K	H C	C C
Control	248	180	167	269	343	323	269	385
Sulfonylurea	62	73	120	151	189	235	209	340
Ca aspirin	123	169	143		255	347	233	
Sulfonylurea + Ca aspirin	*94*	*45*	*46*	*22*	*203*	*297*	*129*	*96*

acting like or by means of insulin FIGURE 6 and TABLE 3 illustrate how salicylates sometimes can enhance the hypoglycemic effect of the sulfonylureas The serum level of salicylates necessary to achieve this action is no higher than 10 mg /100 ml in some of the cases we studied

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ONE YEAR OF CLINICAL EXPERIENCE WITH CHLORPROPAMIDE IN MORE THAN ONE HUNDRED DIABETIC PATIENTS

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During the past 13 months we have treated 130 diabetic patients with chlorpropamide the duration of treatment varying from 1 to 13 months and averaging 6 3 months

Method

Our only criterion for selection of patients for a trial of chlorpropamide therapy was the presence of diabetes in a reasonably cooperative patient The presence of common diabetic vascular complications did not preclude a trial of this drug unless they were acute nor did the occurrence of vascular complications result in a change in therapy as long as the patient remained able to take the drug Chlorpropamide therapy was never begun while the patient was clinically acidotic however the drug was tried later on several patients who were acidotic when first seen The majority of the patients were hospitalized for initial therapy

Most patients were started on the following therapy with minor variations in individual cases If the patient was on a long acting insulin this was discontinued abruptly and he was started on 0 5 gm of chlorpropamide daily usually in divided doses During the initial period of treatment the more acute cases were given regular insulin as indicated by glycosuria Revisions of chlorpropamide dosage were made upward or downward as necessary 5 to 7 days usually being allowed before revision upward due to the accumulation noted in the use of this drug The dosage was gradually increased to tolerance or adequate control this being normoglycemia 75 per cent of the time and the absence of significant symptoms or side effects

We have attempted combined therapy on but one patient No attempt has been made to evaluate this form of treatment

Urine analysis, liver function and hematological studies as well as blood sugar determinations were followed frequently during the initial treatment and approximately once monthly on chronic treatment

Results

Before analyzing our results it is necessary to comment on the patients who compose our series Approximately two thirds of our patients were inmates of a state mental institution As a group these patients constitute a poor risk for any study There are a number of reasons for this including advanced age the average age of our entire series being 61 1 years Most of these patients have mental deterioration that seriously limits their ability to cooperate in such matters as diet The mental changes in many of these patients have been due to previous vascular disease of the central nervous

system, renal insufficiency, or uncontrolled diabetes itself Many of these patients have had previous frank episodes of cerebral, myocardial, or peripheral vascular insufficiency

Of 130 patients, 108 (83 per cent) were considered adequately controlled on chlorpropamide, 83 (64 per cent) are still under study In diabetics previously on less than 60 to 80 units of insulin per day there was a small correlation between prior insulin dosage and likelihood of control with this drug alone (TABLE 1) There was a small but significant correlation between

TABLE 1
CORRELATION OF PRIOR TREATMENT WITH RESPONSE TO CHLORPROPAMIDE*

Prior therapy	Total cases	Response to chlorpropamide	
		Adequate control	Inadequate control
No treatment	17	17	0
Insulin			
0 to 20 units	23	21	2
21 to 40 units	43	36	7
41 to 60 units	19	16	3
61 to 80 units	9	4	5
Over 80 units	9	6	3
Unknown amount	7	5	2
Tolbutamide			
0 to 1.0 gm	9	9	0
1.5 to 2.0 gm	7	5	2
Over 2.0 gm	3	2	1
Total	146	121	25

* Sixteen patients received both insulin and tolbutamide

duration of diabetes and probability of control After a duration of 10 years the patient was less likely to respond to chlorpropamide with adequate control (TABLE 2) There was a slight but positive correlation between age at the time of onset and probability of control, the older patient being the better candidate (TABLE 3) One hundred and eleven of our patients were above 40 years of age at onset of diabetes, and 86 per cent were adequately controlled Only 2 of our patients were juvenile diabetics, and both of these cases failed to respond

Of 4 patients previously poorly controlled on tolbutamide, 3 showed improved control on chlorpropamide Only 4 patients poorly controlled on chlorpropamide were given a trial of tolbutamide, which also failed All 12 patients previously controlled satisfactorily with tolbutamide were also adequately managed with chlorpropamide

No extensive comparative studies between tolbutamide and chlorpropamide have been undertaken However, the following three observations appear reliable First, chlorpropamide is about 3 times more potent than

tolbutamide on a weight to-weight basis Second hypoglycemia can b produced with chlorpropamide in most patients, this is uncommon wit tolbutamide Finally, the onset of action of chlorpropamide is approx mately 50 per cent slower than with tolbutamide, however, the observe cumulative effect with chlorpropamide is more lasting than with tolbutamid This effect has been noted for as long as 7 days following the discontinuanc of chlorpropamide

TABLE 2
CORRELATION OF DURATION OF DIABETES WITH RESPONSE TO CHLORPROPAMIDE

Duration of diabetes (years)	Total cases	Response to chlorpropamide	
		Adequate control	Inadequate control
0 to 1	20	20	0
1 to 2	21	19	2
3 to 5	42	34	8
6 to 10	36	28	8
11 to 20	9	6	3
20	2	1	1
Total	130	108	22

TABLE 3
CORRELATION OF AGE AT TIME OF ONSET WITH RESPONSE TO CHLORPROPAMIDE

Age at time of onset (years)	Total cases	Response to chlorpropamide	
		Adequate control	Inadequate control
0 to 20	2		2
21 to 40	17	12	5
41 to 80	111	96	15
Total	130	108	22

After initial control there appeared to be a gradual reduction in the dosage required for adequate management It has been possible to discontinue the drug entirely in 5 patients after initial control this could represent spontaneous or induced remission At the time these patients were admitted to the series, treatment in addition to diet appeared to be indicated Four of these patients had been on the following treatments 60 units NPH insulin daily for one year 30 units NPH insulin daily for 6 years, 15 units NPH insu-

lin for 12 years followed by tolbutamide (0 5 gm) for 6 months and tolbut amide (3 gm /day) for 8 months The fifth patient was a newly discovered untreated diabetic These patients will be watched with interest

Forty six per cent of our patients on maintenance therapy require 250 mg or less of chlorpropamide daily Seventeen per cent require 250 to 500 mg /day 33 per cent require 0 6 to 1 0 gm /day and only 4 per cent require more than 1 gm /day

In our series there were no untoward hematological effects several of our patients had mild anemias both prior to and during treatment These conditions did not seem to be affected by the drug Low normal white cell counts were seen with some frequency rarely dropping below this level There was no instance of agranulocytosis One patient had a mild chronic lymphatic leukemia that was present prior to treatment and asymptomatic

There were no significant renal effects Many of the patients had albu minuria both prior to and during treatment Most of these were thought to be manifestations of diabetic nephropathy and were apparently unchanged by therapy

Weakness unrelated to hypoglycemia was a complaint of several patients early in the course of the treatment This was not of such severity as to require discontinuance and subsided in a few days

One patient developed a jaundice that was thought to be induced by chlorpropamide This individual developed a rather severe skin reaction and eosinophilia The drug was discontinued because of the skin reaction jaundice was apparent the next day Laboratory studies suggested primarily obstructive type jaundice Both jaundice and skin reaction cleared within a month after the drug was withdrawn After 3 challenge doses of 0 25 gm /day there was a dermatitis and the drug was discontinued Although it was unsubstantiated we feel this was possibly a hypersensitivity type of hepatic reaction

Three other patients developed jaundice while on chlorpropamide therapy In one patient the drug effect was eliminated by surgical correction of an obstructing lesion this patient has subsequently been maintained on treat ment In another patient the drug effect was eliminated by readministration and continued treatment The third patient developed a jaund ce associated with biliary tract disease and frank acute pancreatitis The drug was dis continued during the period of treatment for pancreatit s The pancreatitis and jaundice cleared completely and the drug was resumed The patient has remained clear for 2 months after chlorpropamide was resumed Tran

forms of treatment

The drug was discontinued in a total of 47 patients Seventeen of the 47 patients had the onset of a terminal illness while under study Autopsy was performed in those cases in which the cause of death was not clinically obv ous No death was thought to have been related to the drug The causes of death were generally those expected in elderly diabetics Nine

were from cerebral vascular thrombosis or hemorrhage, and there were 3 episodes of fatal pulmonary emboli Two deaths were due to myocardial infarction with failure There was 1 death due to each of the following staphylococcic pneumonia with lung abscess, staphylococcic pneumonia, and diabetic nephropathy with uremia Special attention was paid to the liver at autopsy in patients who were on the drug at time of death No histological change was noted

The drug was discontinued in 9 of the 47 patients due to poor control Eight of these patients are labile diabetics and are also poorly controlled on their present insulin regime in spite of our best efforts in that direction Seven patients were lost to follow up These essentially represented patients who manifested psychiatric recovery and were thereby discharged from the institution As previously mentioned, it was possible to discontinue the drug in 5 patients because of apparent remission of the diabetic state Gastrointestinal side effects, especially nausea and vomiting, necessitated discontinuance in 5 patients Several other patients manifested lesser degrees of nausea which gradually subsided with continuation of treatment

Chlorpropamide was discontinued in 4 of the 47 cases due to severe skin reactions that did not subside on antihistaminic treatment and continuation of the drug These reactions subsided on withdrawal of chlorpropamide, and the role of the drug was substantiated by a recurrence with a second course of the drug

In addition to the 130 diabetic patients, 6 nondiabetic chronic schizophrenics were given 0 5 gm chlorpropamide daily 4 days weekly for 6 weeks in limited metabolic studies Four of these patients suffered early morning hypoglycemia in spite of midnight feedings This was the only significant observation No effect on the psychiatric state was noted

Summary and Conclusions

(1) Chlorpropamide has been given to 130 diabetic patients for an average of 6 3 months, 83 per cent of these cases were adequately controlled on this medication

(2) Side effects were those usually noted with drugs of the sulfonamide group and occurred in some 12 per cent, requiring discontinuance in 8 per cent Nausea, vomiting, and skin reactions were the most commonly noted side effects One patient manifested a jaundice possibly caused by hypersensitivity to the drug

(3) Chlorpropamide is a potent hypoglycemic agent in a large percentage of nonjuvenile diabetic patients as well as nondiabetic individuals

(4) We feel that chlorpropamide is a significant addition to our armamentarium in the treatment of diabetes mellitus

THE CORRELATION BETWEEN ORAL DOSAGE, BLOOD LEVELS, AND CLINICAL AND METABOLIC ACTIVITY OF CHLORPROPAMIDE IN THE TREATMENT OF DIABETES MELLITUS*

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Chlorpropamide was administered over a period of 8 months to 60 adult diabetic patients These were all private patients among whom were a considerable number sufficiently reliable in their adherence to diet to be suitable for experimental trials Treatment was combined with insulin and other hypoglycemic agents in 12 of the cases Follow up visits were made at frequent intervals both blood sugar and blood chlorpropamide level determinations being made at each visit The objective of the study was to determine the therapeutic effectiveness of chlorpropamide and to ascertain the relationship between dosage blood levels and clinical action

Method

The patients had previously been treated with diet alone with insulin or with tolbutamide Some had also been treated with carbutamide and DBI As in the case of tolbutamide the drug was used to replace all previous medication where possible In two patients it was supplemented with insulin when necessary

The patients were all on a nonweighed diabetic diet and returned at intervals of 3 days to 1 month The blood sugar level was determined 1 hour after breakfast or lunch on each occasion by a picric acid method that yields results equivalent to those of the Fohn Wu test Blood chlorpropamide levels were determined by a spectrophotometric method †[1] The dose of chlorpropamide was deliberately varied widely in a large number of patients usually over the range of 50 mg to 600 mg /day but in a few instances up to 1 5 gm /day The total amount was administered as a single dose after the evening meal In some instances the drug was omitted for trial periods

Of the 60 patients 34 had enough multiple determinations of blood chlorpropamide levels to form the basis for an analysis of the relationship between oral dosage and blood level of the drug In all, 214 determinations were obtained for this purpose, each after at least one week or more at a given dosage level

A composite curve of all the determinations (FIGURE 1) shows a direct proportion between dosage and blood level of chlorpropamide The regression curve shows that for the range of dosage between 50 mg and 1000 mg /day each 100 mg increase in dosage of the drug raises the blood level

* The work reported in this paper was supported by the Clinical Research Department Chas Pfizer & Co Inc Brooklyn N Y

† Developed by the Chemical Research Department of Chas Pfizer & Co, Inc

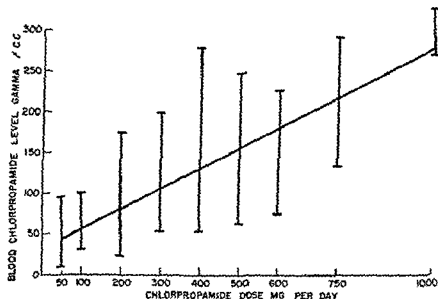


FIGURE 1 Regression curve of relationship between dose and blood levels of chlorpropamide 34 diabetic patients 214 determinations

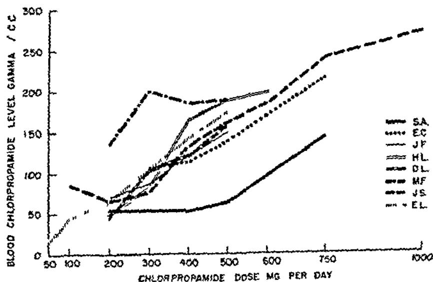


FIGURE 2 Individual patient blood chlorpropamide responses to oral therapy 8 patients at 4 dosage levels

by roughly 25 gamma/cc In FIGURE 2, where the individual responses for

blood chlorpropamide responses

The variability of the blood chlorpropamide levels was analyzed in 10 patients with 14 different patient dosage levels The doses studied varied

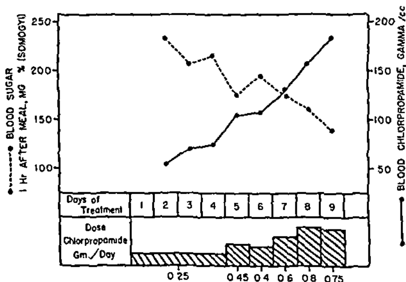


FIGURE 3 Diabetic patient L D acute effect of increasing dosage of chlorpropamide upon blood sugar and chlorpropamide levels over a 9 day period

from 0 2 to 1 0 gm /day, and there were 4 to 10 blood determinations at each dosage level over a period of 1 to 5 months The variability of blood chlorpropamide levels between patients was obviously greater than those for a given patient at a given dosage level In the latter the blood levels varied over only a limited range for long periods of time

The wide variation in dosage of chlorpropamide administered to some 25 diabetics made possible an analysis of the relationship between the dose of drug and pharmacological and clinical effect This was done especially for the purpose of determining the area of dose dependency of the drug The diabetics studied were usually those of moderate severity (requiring up to 40 units of insulin daily), in whom the needed insulin could be replaced completely by chlorpropamide

FIGURE 3 illustrates a subacute experiment A 64 year old white female (L D) with diabetes of 10 years duration had previously been regulated on tolbutamide alone (1 0 gm /day) Under hospital conditions, with fixed

diabetic diet, chlorpropamide was given in increasing doses over a 9 day period. There is a striking effect on blood sugar, with rising dose and blood level of chlorpropamide. While such an acute experiment is interesting, slower increments of dosage are necessary to establish quantitative relationships between dose, blood level, and hypoglycemic effectiveness.

Accordingly, the remaining patients were studied over periods of months with slow stepwise changes in the dose of the drug. As in the case of the other sulfonylureas, the end results of replacement therapy in diabetics can be scored in terms of the blood sugar level achieved one hour after a meal as

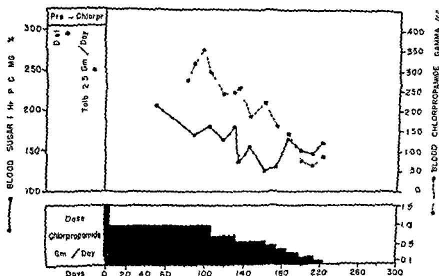


FIGURE 4. Diabetic patient J. S.: a long term study, uniformly good therapeutic effect over a wide range of successively decreasing dosage.

follows: fair or partial regulation, 190 to 250 mg. per cent (Folin Wu), good, 150 to 190 mg. per cent, and excellent, below that level.

Even in the less responsive patients with an end result of only partial regulation the maximum effective dose is still no greater than [illegible] 6 gm. day with a corresponding average [illegible] f 150 gamma per cc. [illegible]
[illegible] with good regulation
[illegible] at the higher levels, although symptoms
o[illegible] were produced, as discussed below.

In the more responsive diabetics increase in dosage over a range of 50 mg. up to 0.6 gm. day may or may not improve regulation from good to excellent. When such improvement did occur it frequently could be correlated with a detectable level of dose and blood level of chlorpropamide. The reverse

could be demonstrated on the reduction of dosage level from the effective levels

In some of the most responsive patients not necessarily the mildest in

In several such cases the drug was omitted and the resultant rise in blood sugar affirmed the activity of such small doses

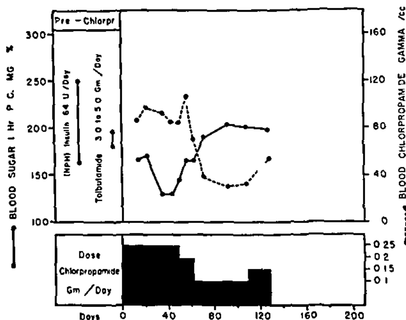


FIGURE 5 Diabetic patient B G a long term study successful replacement of 64 units of insulin and clinical titration of minimum effective dose and blood level of chlorpropamide

The following cases illustrate these principles

Case 1 Patient J S (FIGURE 4) a 33 year old white male with diabetes of 7 years duration He had only a partial effect on 2 5 gm of tolbutamide per day (blood sugar 250 mg per cent Folin Wu 1 hour after meals) On chlorpropamide he had a definitely better effect The inordinately large initial dose did not produce hypoglycemia although it did produce lethargy and other symptoms Decrease in dose and blood levels of chlorpropamide was accompanied by no decrease in effect on blood sugar Indeed there is a suggestion of increasing effectiveness with time in the face of decreasing

in better regulation Even better regulation was achieved on chlorpropamide (0 25 gm /day) Titration by very gradual reduction in dose showed a definite rise in blood sugar at a dosage level of 0 1 gm /day at a blood chlorpropamide level below 70 gamma/cc The effective replacement of so large a dose of insulin by so small a dose of sulfonylurea is striking

Case 3 Patient F B (FIGURE 6) a 79 year old female diabetic with diabetes discovered *April 1958 with an initial blood sugar of 350 mg per cent* Tolbutamide (2 0 gm /day) produced an excellent therapeutic response

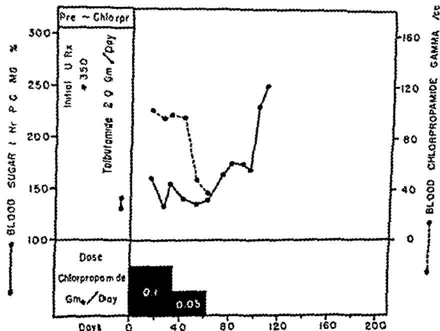


FIGURE 6 Diabetic patient F B a long term study low dosage and blood level requirement of chlorpropamide in mild diabetes proof of effectiveness by withdrawal of drug

Chlorpropamide therapy was begun in May 1958 with an initial dose of 0 1 gm /day The response was maintained on reduction of dose to 50 mg /day and a blood chlorpropamide level of 28 gamma/cc Omission of all therapy resulted in a gradual rise in blood sugar to 250 mg per cent over the next 40 days

The dose dependency of chlorpropamide noted in patients studied with total replacement of insulin could also be ascertained by titration in severe diabetics in whom insulin could not be entirely omitted In one such instance chlorpropamide (0 4 gm /day) replaced 5 units of insulin at a blood level of 159 gamma/cc while 0 6 gm /day replaced 11 units of insulin at a blood level of 210 gamma/cc

Of the 60 patients studied several were given excessively large doses deliberately or because of the exploratory nature of the early therapeutic studies Nine patients had definite symptoms of overdosage consisting of subjective muscular weakness a groggy or fuzzy feeling lassitude numbness and tingling in toes and nausea All managed to perform their mental tasks although with an overlay of lassitude and sleepiness Ataxia occurred in one exceptional individual who received 3 0 gm /day

The daily dosage levels associated with such symptoms were 1 patient 3 gm 2 patients 1 5 gm 3 patients 1 0 gm 1 patient, 0 75 gm and 1 patient 0 4 gm There was a definite correlation with blood chlorpropamide level which was usually over 250 to 275 gamma/cc at the time of symptoms This was underscored by the exceptional patient who had symptoms at a dosage level of 0 4 gm /day while due to unusually slow disposal the blood level was 280 gamma/cc However other patients were asymptomatic at blood levels of 275 to 295 gamma cc as were others on doses of 0 75 to 1 0 gm /day This illustrates certain individual factors involved in the subjective symptoms of overdosage

All symptoms of overdosage disappeared readily within 4 to 5 days or less of reduction in dosage even in the most highly overdosed individual (3 0 gm /day) with no residual symptoms

Finally the average and usual dose of definitive regulation of all these cases was 0 1 to 0 4 gm day so that the 0 75 to 1 5 gm dose productive of symptoms was 2 to 5 times the therapeutically necessary dose

Discussion

It is of considerable theoretical importance that there is a proportionality between oral dosage and blood levels (and presumably hypoglycemic activity) of chlorpropamide in treated diabetics this is especially true in individual patients The relationship is closest with least variability in blood chlorpropamide levels in individual patients on a given dose of chlorpropamide who may show remarkably constant blood levels for several weeks at a time No accumulation of drug in the blood occurs even after months of therapy

Since patients vary widely in the dose required for optimal regulation this leads to the distinct clinical advantage that the drug may be administered in varying doses without determination of blood levels However individual patients do show variations from one another in their blood response This helps to explain unexpected treatment failures and suggests the judicious elevation of the dose of chlorpropamide above the usual maximum (0 6 gm /day) in such patients

The range of effective blood levels is quite wide from 25 to 175 gamma/cc The effectiveness at the low dosage and blood levels in responsive patients is striking

While clinical experience suggests that the other sulfonylureas (carbutamide and tolbutamide) probably also have the same dose—blood sulfonylurea response relationship this type of extensive long term study has not been performed for those drugs

The risk of hypoglycemia is small in spite of the great clinical effectiveness of chlorpropamide. The level of overdosage is far beyond the usual therapeutic range, and its symptoms are so mild and easily recognizable as to present no clinical problems. It is of interest that one symptom, muscular weakness of the lower extremities, has been produced in the experimental animal by very large intravenous doses of tolbutamide (270 mg./kg./hour), and is due to direct action on the spinal cord.[2]

Summary

Chlorpropamide has been administered to 60 diabetics over a period of 8 months. Both clinical response and blood chlorpropamide levels were measured. In general, there is a close relationship between dose and blood chlorpropamide response, but there are some individual variations among patients in such response. Patients maintained blood chlorpropamide levels within narrow limits at a given dosage level over long periods of time. There is no accumulation of the drug after long periods of therapy. *The routine determination of blood chlorpropamide is unnecessary in clinical regulation of diabetics, although it may be useful in some nonresponsive patients.*

The effective dosage range in the treatment of diabetics is 50 mg. to 0.6 gm./day, and the corresponding blood levels are 25 to 175 gamma/cc. In some patients a demonstrable level of blood chlorpropamide for effectiveness may be determined. Hypoglycemia is rare, and symptoms of overdosage are mainly mild weakness. This does not usually occur below a dose of 0.75 to 1.0 gm./day, which is beyond the usual therapeutic range.

Acknowledgment

The statistical analysis of the data used in the preparation of FIGURE 1 was performed by Hilda Simon of the Statistical Department of Chas. Pfizer & Co., Inc.

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CLINICAL EXPERIENCES WITH CHLORPROPAMIDE, WITH SPECIAL REFERENCE TO COMPARISON WITH PREVIOUS EXPERIENCE WITH CARBUTAMIDE AND TOLBUTAMIDE

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At this writing it is about 3 years since we first used sulfonylurea preparations in Germany for the treatment of diabetics During this period we have employed these drugs systematically and on a large scale A short time ago I reported our accumulated experience with these new therapeutic agents In that summary I emphasized the fact that there can no longer be any doubt that oral sulfonylurea therapy has proved to be of value in the treatment of diabetes despite the fact that the mechanism of action of these substances remains to be clarified completely Approximately 40 per cent of all diabetics can be treated successfully by means of oral administration of these antidiabetic drugs, and the metabolism can be stabilized satisfactorily for the duration of the therapy The number of "late failures" did not exceed 10 per cent The incidence of undesirable side reactions was 8 per cent in the case of carbutamide (BZ 55) and less than 1 per cent in the case of tolbutamide (D 860)

Carbutamide and tolbutamide are the 2 substances that heretofore have been generally available in Germany for oral sulfonylurea therapy Preference has been given to the latter, both in Germany and, to the best of my knowledge, in the United States also, because it does not possess sulfonamide characteristics Therefore it is significantly better tolerated and exhibits an incidence of untoward side reactions considerably smaller than that of carbutamide

Recently, experimental and clinical studies have been conducted in the United States on chlorpropamide, a new sulfonylurea derivative We have been the first investigators to use this new antidiabetic drug in Germany, employing it for the treatment of diabetics in the Diabetic Pavilion of the Waldkrankenhaus We had available information supplied by the manufacturer concerning the physical and chemical properties of chlorpropamide, its intensive hypoglycemic action in the dog and rat, and the observed lack of damage to the internal organs of these test animals

Materials and Methods

Altogether, we have completed control with chlorpropamide in 50 hospitalized diabetics In an additional 24 cases, full metabolism adjustment has not yet been completed While this is an admittedly rather small test group, the patients were very carefully examined and specific tests were conducted to determine the effect of therapy on carbohydrate metabolism and to detect any untoward effect which might develop All patients received thorough examinations at intervals of several weeks, beginning with

the initiation of treatment I therefore believe that we can offer cer valid conclusions on the basis of our experiences

TABLE 1 presents the composition of our test group according to age may be seen, the 50- to 60 year age group clearly predominates with patients Eleven other patients were between 40 and 50 years of age Germany we classify adult diabetics (those having maturity-onset diabe chiefly into 2 categories first, the sthenic type, which includes patients v

TABLE 1
AGE GROUPS OF THE DIABETICS TREATED WITH CHLORPROPAMIDE

Age (years)	Men	Women	Total
21 30	2	1	3
31–40	—	—	—
41 50	4	7	11
51 60	12	11	23
61 70	1	6	7
71–80	1	5	6
	20	30	50

TABLE 2
DURATION OF THE DIABETES BEFORE INITIATION OF CHLORPROPAMIDE THERAPY

	Newly diagnosed	Time (years)								
		½	1	1½	2	3	4	5	7	More than 7
Number of patients	10	3	4	3	7	6	6	6	1	4

appear well nourished and in whom insulin output may be considered to b normal or near normal, but who develop the disease late in life due to th effects of other endocrine glands on the metabolism of the patient, anc second, the asthenic type, which includes patients who are thin, not we nourished, who suffer easily from fatigue, and whose insulin output may b considered to be deficient

It is the sthenic type of diabetic who generally responds to therapy wit oral antidiabetic agents, the asthenic type usually is not amenable to suc therapy The majority of our patients were of the sthenic type, and only could be considered as definitely of the asthenic type

TABLE 2 indicates the duration of diabetes in each individual case, a shown, the minimum duration is six months, the maximum more than seve years It would appear, however, that the duration of the disease is not a important criterion for determining the advisability of initiating sulfonylure

therapy This is the same impression we gained from evaluation of the other two aforementioned antidiabetic agents

What does appear to be more important is the age at which the disease first manifests itself We already knew from our experiences with carbutamide and tolbutamide that, in general, the prospects for success were better the later diabetes manifested itself after the fortieth year TABLE 3 shows this relationship in terms of our patients treated successfully with chlorpropamide three of them were cases where the disease developed before the

TABLE 3
AGE AT ONSET OF DIABETES IN PATIENTS SUCCESSFULLY TREATED WITH CHLORPROPAMIDE

	Age (years)																							
	26	36	39	42	43	44	45	46	47	48	49	50	51	52	54	56	57	58	59	60	63	64	65	67
Men	1	—	—	1	1	1	3	1	1	1	—	2	1	1	—	1	2	1	1	—	—	1	—	—
Women	—	1	1	—	1	—	—	3	—	—	3	1	—	—	1	—	1	—	—	1	3	1	1	1

TABLE 4
PREVIOUS TREATMENT OF THE DIABETICS WHO RESPONDED SUCCESSFULLY TO CHLORPROPAMIDE

No treatment	9
D et alone	7
Diet and insul n	8
Diet and carbutamide	5
Diet and tolbutamide	19
Diet tolbutam de and insul n	1
D et and guabeta	1

patient s fortieth year This is particularly worthy of attention The rest of the patients are distributed regularly with respect to the first appearance of the disease between the forty second year and the sixty seventh year

TABLE 4 shows that some of our patients had not been treated before initiation of chlorpropamide therapy In these cases the diabetes has been diagnosed very recently In others treatment consisted of dietary measures either alone or with insulin A third group had received either carbutamide or tolbut
This last group
opportunity for

Results

On the basis of
propamide appe
namely the sther

asthenic type in whom diabetes is a result of a pronounced lack of insulin, chlorpropamide seems to be just as unsuitable as the other two sulfonylurea preparations. Of course there are exceptions to this general rule; indeed, the treatment of all diabetics must be so carefully based on each individual case that one can say there is hardly any other disease that requires such strict individual attention. Aside from this, we had the impression in a few other cases that the use of chlorpropamide was possible in cases where carbutamide and tolbutamide had previously failed to bring about metabolic compensation at the same dosage levels. Two cases illustrating this point are shown in

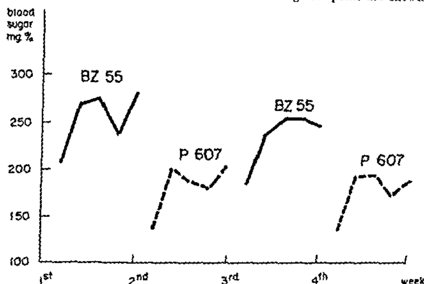


FIGURE 1. Blood sugar levels in relation to treatment with chlorpropamide and carbutamide. Patient F. W. H. male 53 years old. Dosage was 1 tablet twice daily (1000 mg daily).

FIGURES 1 and 2. In FIGURE 1, one carbutamide tablet twice daily, for example, was insufficient to maintain the metabolism in balance; the daily blood sugar profile showed values as high as 280 mg. per cent. In contrast, 1 chlorpropamide tablet twice daily maintained blood sugar values at considerably lower levels, never exceeding 200 mg. per cent. FIGURE 2 shows a similar situation in regard to tolbutamide as compared with chlorpropamide therapy.

I should also like to call your attention to 2 other points. First, when equivalent dosage of the antidiabetic agents is administered, the hypoglycemic action of chlorpropamide is definitely more potent than that of tolbutamide. Second, the fasting blood sugar, especially, is lower when chlorpropamide is administered. We observed these effects quite frequently in our test patients, and thereby we were able to come to important conclusions regarding dosage for chlorpropamide.

At the beginning of our tests we generally used an initial dosage of 500 mg. of chlorpropamide three times daily. This dosage was never exceeded, and

we continued to administer it until a definite improvement in metabolic compensation was evident, with urine sugar and blood sugar values approaching normal As our experience accumulated, we learned that for a number of patients an initial dose of 500 mg twice daily would achieve the desired result and, in a few cases, we were able to initiate therapy with a single daily dose of 500 mg of chlorpropamide This was possible, however, in only a small proportion of the total cases Furthermore, the initial dosage, as well as subsequent maintenance dosage, required adjustment to individual need,

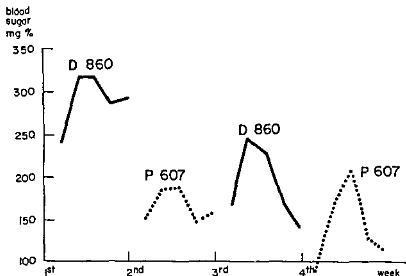


FIGURE 2 Blood sugar levels in relation to treatment with chlorpropamide and tolbutamide Patient M W, female, 57 years old Dosage was 1 tablet twice daily (1000 mg daily)

to the individual response of each patient, to the tempo at which the compensation of the metabolism progressed, and to other such factors Thus, as with all antidiabetic medication, over all indications can be given for dosage, but no hard and fast precise directions can apply to all individuals In a number of our patients we observed that satisfactory metabolic compensation could be obtained only if the initial dosage was 500 mg three times a day administered for several days

With occurrence of satisfactory metabolic compensation the initial dosage was gradually reduced after 4 to 6 days, being replaced by a maintenance dosage of 125 to 1000 mg /day, as shown in TABLE 5 Few patients required a daily maintenance dosage as high as 1000 mg , and these were mostly cases whose sugar metabolism had not been maintained in balance by the same dosage of carbutamide or tolbutamide In some of these cases examination a few weeks later revealed that metabolic compensation had improved to such an extent that the daily maintenance dosage could be reduced to 500 mg

or in some cases even to 250 mg According to my recollection this is an observation we scarcely ever encountered during our earlier *investigation of tolbutamide* With some reservations one can perhaps conclude from this observation *that under chlorpropamide medication the poorly functioning pancreas improves somewhat and again begins to produce insulin in* more adequate amounts

From the facts I have already presented it is clear that it is still somewhat premature to make any definitive statement concerning the maximal initial and maintenance dosages of chlorpropamide As I have stressed this must *be a matter of individual adjustment* Nevertheless we do have the impression that the maintenance dosage *is frequently considerably lower than with*

TABLE 5
MAINTENANCE DOSE OF CHLORPROPAMIDE

	Chlorpropamide (mg)					
	1000	500	250	250 every second day	125	125 every second day
Number of patients	9	14	13	1	6	1

carbutamide and tolbutamide so that in many cases the sugar metabolism can be balanced satisfactorily as long as chlorpropamide maintenance dosages on the order of 125 250 or 500 mg are continued Only conjectures can be offered for explanation of this behavior *Chlorpropamide is absorbed very quickly from the gastrointestinal tract On the other hand the substance is eliminated from the body very slowly Because of these factors it is possible* to preserve a blood level of chlorpropamide high enough to accomplish a satisfactory hypoglycemic action with very small daily dosage It is also possible that the explanation of the more potent hypoglycemic effect of chlorpropamide lies in the same factors All of these questions require further investigation *As I have already indicated we were impressed throughout our study by the fact that the fasting blood sugar frequently was quite low* For this reason we have preferred to administer the drug early in the day and according to patient need giving either a daily dosage of one half or one full tablet immediately after breakfast as a single dose or where daily dosage is required of 1 tablet twice daily that is, after breakfast and after lunch

Patients *previously treated with insulin can be successfully placed on chlorpropamide therapy* just as is the case with carbutamide and tolbutamide With such patients we were able to replace daily insulin dosage as high as 48 units with chlorpropamide One of these cases is shown in FIGURE 3 Heretofore the patient had taken 32 units of insulin daily *Five hundred mg* of chlorpropamide daily did not produce a complete *normalization* of the blood sugar values which we *consider* the necessary criterion for the lasting

success of oral diabetic treatment With 2 daily doses of 500 mg each the blood sugar was completely normalized so that the use of insulin could be discontinued It is worthy of mention that in a number of newly diagnosed diabetics who suffered from acidosis before initiation of treatment we did not use insulin at all but tested the effectiveness of chlorpropamide by administering it from the beginning as the only antidiabetic therapy given The acidosis d sappeared and the metabolism was normalized This was a most interesting observation but it was merely in the nature of a clinical

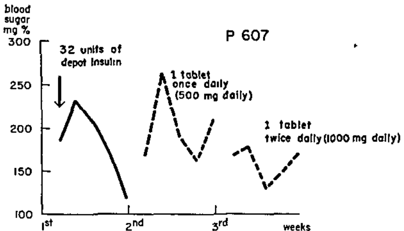


FIGURE 3 Blood sugar levels n relat on to treatment with chlorpropamide and nsul n Patient W S male 52 years old Dosage was as shown

experiment and I do not recommend it as a regular procedure to be followed

returned for examination and it was found that in both cases the metabolism was decompensated Even when the dosage was increased to 500 mg three t mes daily there was no improvement so that it was necessary to reinstitute insulin therapy

Side Effects

We devoted particular attention to the question of whether chlorpropamide had any deleterious side reactions such as may be seen to some degree with carbutamide The following may be reported up to a dosage of 500 mg

following an attack of hepatitis Serum bilirubin amounted to 15 mg per

cent and tests of liver function gave abnormal results as expected Heretofore the patient had been taking 40 units of insulin daily, which we replaced

chlorpropamide dose could be reduced to 250 mg After an additional month and one half of therapy examination of the liver showed completely normal results the liver was no longer enlarged and the function tests gave normal results The patient felt healthy and had gained weight This case shows not only that hepatic damage need not be feared with chlorpropamide but also that existing hepatic damage can heal during chlorpropamide treatment

The appearance of a pruritic measleslike exanthema was noted in three patients some four to six weeks after initiation of medication with chlorpropamide therapy This side reaction disappeared a few days after the drug was discontinued Examination of metabolism indicated that further administration of chlorpropamide was no longer necessary, since metabolic balance could now be maintained by dietary means alone A so called thrust effect of the sulfonylurea substances occurred, we had seen this previously when using carbutamide and tolbutamide Furthermore, in these cases there did not appear to be a drug allergy to chlorpropamide, since the intracutaneous tests and the Prausnitz Kustner test were negative

Using an initial dosage of 500 mg three times daily we observed a number of cases of gastrointestinal disturbances principally epigastric distress, feelings of pressure in the stomach and nausea For the most part however these disappeared when the dosage was decreased so that maintenance dosages of 250 to 500 mg /day were only rarely accompanied by epigastric manifestations Interesting in this connection is the reaction of a few patients who had also complained of epigastric distress when carbutamide and tolbutamide were used When these patients received the smaller, but fully effective, dosage of chlorpropamide, they no longer complained of epigastric distress Apparently they tolerated a therapeutic amount of chlorpropamide better than they did either of the other 2 sulfonylurea preparations

Conclusion

Again I stress most emphatically the fact that our clinical investigation of chlorpropamide is preliminary in nature and involves tentative tests conducted with a relatively small group of test patients, hence it is not feasible to come to final and definitive conclusions from this work Further research should be conducted to establish validity in regard to indications, tolerance side reactions and toxicity In future investigations it will be important to explore further possible side effects that may involve the hepatic or hematopoietic systems As I have indicated, we have never seen any sign of such effects but other investigators have reported occasional occurrences Therefore further evaluation of this new oral antidiabetic agent is necessary

CLINICAL EXPERIENCES WITH CHLORPROPAMIDE GIVEN TO HOSPITALIZED PATIENTS

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Introduction

Oral therapy for diabetes is no longer a novelty As months have grown into years of experience with oral sulfonylurea compounds the apprehension that harm might ensue from their use over long periods is subsiding As a result search for the most effective compound with the least toxicity has been intensified This paper deals with a clinical testing of chlorpropamide in 15 hospitalized diabetic patients with a follow up of 6 months The duration of the diabetes ranged from 1 week to 16 years (TABLE 1) Seven patients received a daily average dose of 30 U of insulin prior to the test period One was taking 55 U daily Two patients were receiving tolbutamide with unsatisfactory results In 6 patients the diabetes was not under good control none of these was receiving insulin or tolbutamide

Ideally the patients for chlorpropamide therapy would be selected from approximately 10 per cent of the diabetic population This is based on the relatively sound assumptions that it is possible to control diabetes barring acute complications in 80 per cent by diet alone that 5 per cent are children and that the remaining 5 per cent are adults with labile diabetes In these two latter groups long term treatment with chlorpropamide is ineffective

Method

A few preliminary days of observation preceded the test period that consisted of a minimum of 3 consecutive 5 day periods During the first 5 days the amount of insulin given was reduced or eliminated and in the case of patients receiving tolbutamide this drug was withdrawn In each instance the object was to permit hyperglycemia and glycosuria to develop in order to discern the effect of chlorpropamide more clearly During the second 5 day period chlorpropamide was administered and throughout the third period this drug was omitted Throughout the 15 days daily blood sugar determinations were made by the Somogyi Nelson method on samples taken in the fasting state and at 3 P M with daily quantitative determinations of the glycosuria Tests for acetonuria were performed on successive voidings when the insulin therapy was reduced Other laboratory studies were made during the first and toward the end of the second week The diet remained constant throughout and patients were guarded against undernutrition

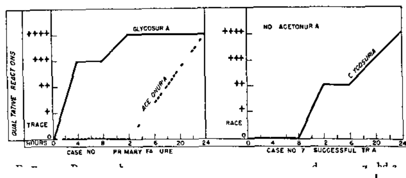
* Research Fellows in Metabolism

TABLE 1
DATA ON CHLORPROPAMIDE THERAPY AND RESULTS IN 10 CASES OF FAVORABLE RESPONSES

Case	Patient age color sex	Duration of diabetes	Prior treatment	Percentage fall			Chlorpropamide daily (gm)	Other
				Blood sugar		24 Hour quantity in urine		
				Fasting	3 P M			
1	MC 66WF	1 month	Insulin 10 U	56 3	41 5	—	0 5	
2	JR 42WM	7 years	Diet	45 5	33 7	69 7	0 5	Tolbutamide failure
3	AM 79WM	1 week	Insulin 8 U	47 1	51 2	37 5	0 5	
4	CS 60WF	2 weeks	Insulin 15 U	74 1	58 2	92 3	0 25	Chlorpropamide 0 5 gm daily
5	CR 73WF	6 months	Tolbutamide 1 5 gm	52 6	14 8	71 4	1 0	Tolbutamide failure
6	GR 76CM	6 months	Tolbutamide 1 5 gm	57 1	40 1	88 5	1 0	Tolbutamide fair control
7	BS 57WF	16 years	Insulin 40 U	60 5	54 9	91 5	1 0	
8	LB 68WM	16 years	Insulin 36 U	33 0	27 4	99 8	0 75	Tolbutamide failure
9	EJ 85WF	6 years	Insulin 44 U	44 1	24 4	—	0 75	
10	JC 51CF	2 weeks	Insulin 55 U	4 2	11 6	46 2	1 0	
Range		1 week to 16 years	Insulin 8 to 55 U	4 2 to 74 1	11 6 to 58 2	37 5 to 98 8	0 25 to 1 0	
Average		4 6 years	Insulin 30 U	47 5	35 8	87 0	0 73	

Results

none of these was adequate control of the diabetes ac ... as necessary One patient who ... of tolbutamide daily had fasting ... cent with an average of 296 mg ... prompt reduction of the fasting blood sugar to 82 mg per cent The comparative effects of *tolbutamide and* chlorpropamide as observed during the period of hospitalization indicate (TABLE 2) that chlorpropamide in this instance is a *more potent blood sugar* reducing agent than tolbutamide This impression was supported by a



In 2 cases in which tolbutamide had been a recent secondary failure excellent control of the diabetes was achieved with chlorpropamide The drug had a blood sugar lowering effect in 10 of the 15 patients

Following discharge from the hospital of these 10 patients the diabetes in each of them has been controlled by 0 25 to 1 0 gm of chlorpropamide daily The average amount given in 1 dose daily was 0 75 gm Experience with this drug and hypoglycemic reactions caused by it have prompted us to begin therapy with a daily dose of 0 5 gm and modify it as needed Addi

TABLE 2
EFFECTS OF IDENTICAL DOSES OF CHLORPROPAMIDE AND TOLBUTAMIDE ON THE BLOOD SUGAR VALUES AND ON THE DEGREE OF GLYCOSURIA IN ONE PATIENT

	Average			6 Month follow up average & range blood sugars mg per cent
	FBS mg per cent	3 P M Blood sugar—mg per cent	Urine sugar gm /24 hour	
Pretest control	228	284	9 8	
Chlorpropamide 1 0 gm	147	168	2 1	194 (147 210)
Tolbutamide 1 0 gm	184	262	11 3	296 (271 342)

tions of more than 0 25 gm to the daily dose in 1 week increased the risk of hypoglycemic reactions

A typical favorable response to chl
FIGURE 2 Both fasting and
rected Furthermore followir
of the blood sugar to abnormally high levels ensued

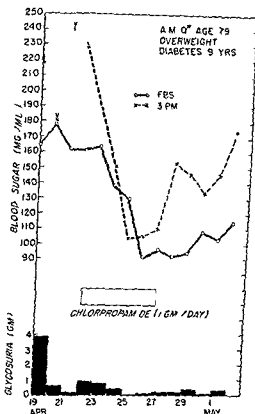


FIGURE 2 The hyperglycemia and glycosuria observed in the preliminary control period were corrected by 1 0 gm of chlorpropamide daily The favorable effect gradually subsided after the chlorpropamide was omitted

Primary Failures

Five primary failures with chlorpropamide therapy were noted In 2 cases acetonuria appeared in increasing amounts in the first 24 hours after the withdrawal of insulin, as illustrated in FIGURE 1 In another case an acute cholecystitis with cholelithiasis was a complicating factor, in, 1 case metastatic carcinoma was present, there was 1 patient who had gangrene of an extremity

Data in the case of a failure to respond favorably to chlorpropamide are

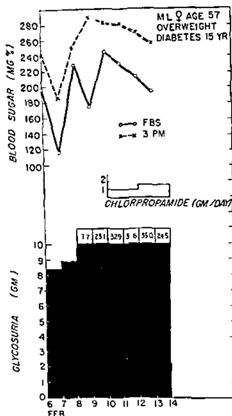


FIGURE 3 The hyperglycemia and glycosuria were not significantly improved in this case during five days of chlorpropamide therapy

shown in FIGURE 3 Neither the hyperglycemia nor the degree of glycosuria was favorably influenced by the drug

Side Reactions

Hypoglycemic reactions, mild in nature were accompanied in two cases

0 25 gm prevented hypoglycemic reactions

A transient pruritic, maculopapular cutaneous eruption occurred on 1 fore arm of 1 patient who had received 1 gm daily of the drug for 1 month The rash receded under influence of antihistamine therapy and there has been no recurrence without interruption of chlorpropamide therapy

Special Tests

An abnormal alkaline phosphatase value was noted in one instance, but this was doubtless due to right sided heart failure with auricular fibrillation This patient died 1 month after these studies were completed The liver exhibited a marked central vein congestion considered characteristic of the cardiac disease, with no other detectable abnormalities In none of the remaining 14 patients was there any detectable disturbance in the hematopoietic, renal or hepatic functions

Acute Effect of a Single Dose of Chlorpropamide

The speed of action, degree of effect and the duration of the blood sugar lowering effect of a single dose of chlorpropamide were observed when a liquid dietary formula comprised of protein, 113 gm , carbohydrate, 268 gm , and fat, 63 gm (2090 calories) was divided into 12 equal amounts, 1 of which was

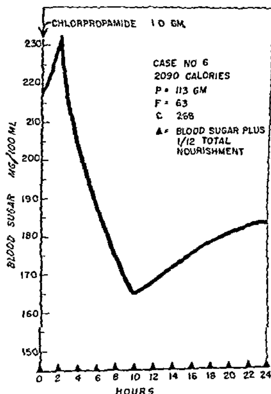


FIGURE 4 The effect of a single dose (1 gm) of chlorpropamide is illustrated Identical liquid nourishments were given at 2 hour intervals for 24 hours The blood sugar lowering effect became apparent between 2 and 4 hours after the first nourishment was given This effect was most marked between 8 and 12 hours, but was still apparent after 24 hours had elapsed

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10 hours after which a slow and very gradual increase begin A marked depressing effect on the level of the blood sugar was still manifest 24 hours after the drug had been administered

The effect of 0 5 gm given twice daily is compared in TABLE 3 with that of 1 gm given in 1 dose The single dose produced the more consistent

TABLE 3

DEGREE AND DURATION OF EFFECT OF 0 5 GM OF CHLORPROPAMIDE ON THE FASTING AND POSTPRANDIAL BLOOD SUGAR LEVELS AS COMPARED WITH THOSE FOLLOWING THE ADMINISTRATION OF 1 0 GM GIVEN ONCE DAILY

Chlorpropam de	Average		Urine sugar gm 24 hour
	FBS mg per cent	3 P M B ood sugar mg per cent	
0 5 gm b i d	108	242	2 8
1 0 gm single dose	147	168	2 1

blood sugar lowering effect On this basis because of sim lar observations we give a single dose of chlorpropamide before breakfast daily

It was observed that with the daily administration of chlorpropamide its apparent effect increased for several days after which a plateau of effect ensued Also after withdrawal of the drug the blood sugar lowering effect abated gradually over a 3 to 5 day period

No instance of decreasing effect of chlorpropamide during therapy up to and in excess of six months has been observed It is possible and indeed probable that the lack of secondary failures is due to the brevity of the per od of therapy

Summary

This study of the properties of chlorpropamide involving 15 closely observed hospitalized patients indicates that this drug is a potent blood sugar reducing agent when given in doses as small as 0 25 to 0 5 gm /day the blood sugar

the d abetes in 10 of the 15 patients having adult acquired diabetes in no

apy, and the detection of acetonuria in increasing amounts after a reduction of the amount of insulin given served as a reliable guide in detecting those patients in whom a satisfactory response would not be obtained whereas, in cases in which no acetonuria occurred there were effective responses to chlorpropamide therapy

Prolonged therapy under close surveillance is still necessary to prove the harmlessness of this drug Also, the likelihood must be guarded against that such an effective agent might receive widespread adoption as a substitute for appropriate dietary measures

OBSERVATIONS ON THE MODE OF ACTION AND CLINICAL EFFECTS OF CHLORPROPAMIDE IN DIABETES MELLITUS

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Preliminary clinical studies of chlorpropamide have demonstrated this drug to be an effective hypoglycemic agent in man In higher dosage ranges

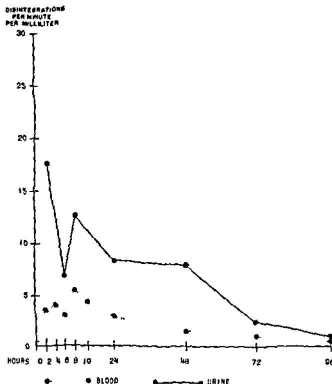


FIGURE 1 Serum and urinary levels after oral administration of 1 0 gm. of S^{35} chlorpropamide

it has produced marked hypoglycemia in susceptible persons It has permitted control of clinical and laboratory abnormalities in mild to moderate

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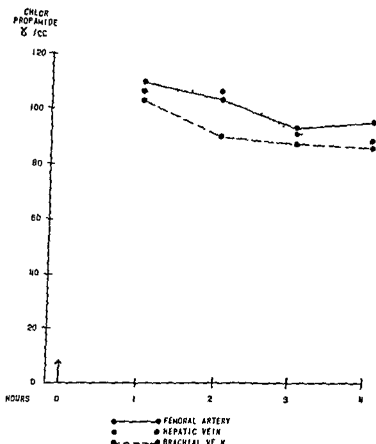


FIGURE 2 Comparison of arterial hepatic venous and peripheral venous levels of chlorpropam de after admin stration of 1 0 gm of the drug intravenously

its distribution and excretion its effects on arterial venous and hepatic vein blood sugar levels its influence on the prednisone glucose-tolerance test and epinephrine induced hyperglycemia and experience with it and other sulfonylurea derivatives on diabetes made overt by liver injury

Materials and Methods

Distribution and excretion were studied in 2 diabetic subjects by measuring urinary output of orally administered S^{35} chlorpropamide and in 10 non diabetic subjects by determining drug levels in arterial venous and hepatic venous blood after oral or intravenous administration of nonradioactive

chlorpropamide. The relationship of drug dosage and serum level to blood sugar changes for a 48 hour period was investigated in 6 patients with diabetes by administering 1.0 gm. of chlorpropamide as a single dose orally. Results of prolonged administration of the drug on its serum concentration were investigated by obtaining weekly determinations in 12 patients receiving 0.5 or 1.0 gm. of chlorpropamide each day. Comparison of arterial, venous and hepatic vein drug and sugar levels was made in 10 patients by using the technique of hepatic vein catheterization.[3] Effects of chlorpropamide on

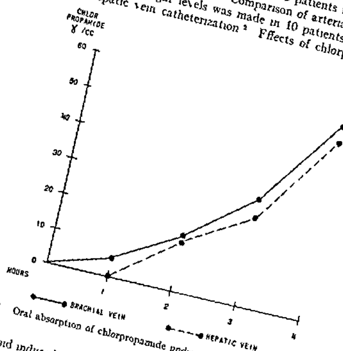


FIGURE 3. Oral absorption of chlorpropamide under condition of hepatic vein catheterization.

glucocorticoid induced hyperglycemia were studied by performing a standard oral or intravenous glucose tolerance test, repeating the test with prior administration of cortisone[a] or prednisone[a] and finally obtaining a glucose tolerance test after receipt of the adrenal steroid plus 1.0 gm. of chlorpropamide. The influence of chlorpropamide on epinephrine induced hyperglycemia was evaluated by studying the effects of 0.01 cc. of a 1:1000 solution of intramuscular epinephrine on blood sugar before and after administration of 0.5 gm. of chlorpropamide each day for 5 days. Blood sugars were determined by the method of Folin and Wu. Chlorpropamide was determined in the serum by an ultraviolet spectrophotometric procedure.[6]

Observations

After oral ingestion of 1.0 gm. of S^{35} chlorpropamide there was a peak rise in serum radioactivity at 4 to 6 hours followed by a gradual decrease in

serum levels and a concomitant increase in urinary radioactivity in the ensuing 48 hour period. Radioactivity was present in the serum and urine 96 hours later (FIGURE 1). A comparison of drug levels in arterial, venous and hepatic venous blood after parenteral administration of chlorpropamide showed a small but definite difference due to its uptake and metabolism by the liver and other tissues and excretion by the kidney (FIGURE 2). After oral administration of the drug in healthy subjects chlorpropamide could be

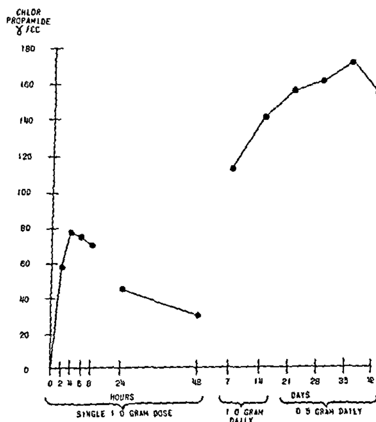


FIGURE 4. Venous levels of chlorpropamide after oral administration of the drug

detected in the pe
presumably becau
hepatic cirrhosis
the presence of intrahepatic and extrahepatic shunts.[2]

Peripheral drug levels varied considerably in different subjects after a single or repeated daily doses of chlorpropamide. A level of 40 gamma or more was usually maintained for 24 hours and was often still present at 48 hours after ingestion of 1.0 gm of the drug (FIGURE 4). Weekly drug levels in 12 ambulatory patients showed a cumulative effect of the drug with

daily administration of 1.0 gm. and a similar pattern in some subjects receiving 0.5 gm. (FIGURE 4).

Blood sugar effects. Simultaneous blood sugar and drug levels after oral administration of chlorpropamide often showed an inverse relationship; however, the sugar response varied markedly and did not appear specifically to depend upon the height of the drug level. Although patients receiving 1.0 gm. of the drug daily often had a progressive increase in serum chlorpropamide and a decrease in blood sugar, there was usually a leveling off of both

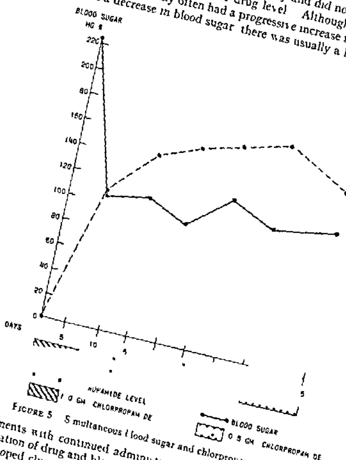


FIGURE 5. Simultaneous blood sugar and chlorpropamide levels.

measurements with continued administration of 0.5 gm. daily (FIGURE 5). No correlation of drug and blood sugar levels was demonstrable in 3 subjects who developed clinical evidence of hypoglycemia. Much higher chlorpropamide levels were noted in subjects who responded to this drug but did not develop hypoglycemia (FIGURE 6).

An earlier and greater hypoglycemic effect was noted when chlorpropamide was administered intravenously. The hepatic venous glucose level usually showed an earlier decrease than the arterial glucose, indicating a primary

hepatic action as previously demonstrated with insulin,[6] phenethyldiguanide,[7] and other sulfonylurea derivatives[8] A rebound increase in hepatic venous sugar was noted and attributed to release of epinephrine[6] While a significant hepatic venous arterial glucose difference was noted, little change occurred in the peripheral arterial venous glucose difference in 6 subjects without liver disease (FIGURE 7) In contrast, 2 of 5 subjects with cirrhosis and evidence of vascular shunts exhibited a definite peripheral effect Simultaneous chlorpropamide and blood sugar studies in 1 of these subjects showed

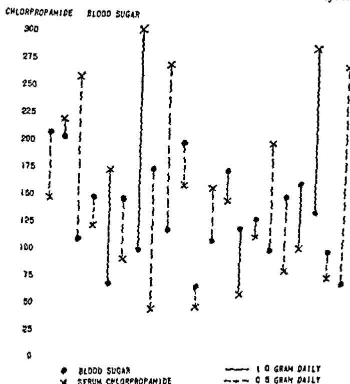


FIGURE 6 Chlorpropamide and blood sugar levels in selected diabetic patients on maintenance therapy

diminished drug uptake by the liver little alteration in hepatic venous glucose, and increased peripheral arteriovenous glucose difference after intravenous injection of 1 0 gm of the drug (FIGURE 8) This phenomenon, which has numerous implications,[9-12] requires further study Its selective nature is probably related to the location and number of shunts and the

hyperglycemia and glycosuria in patients with latent diabetes[3] or liver disease[4] Studies were conducted on the influence of chlorpropamide on carbo

of an adrenal steroid was followed by an abnormal glucose tolerance test reaction) were chosen for study In each instance chlorpropamide blocked anticipated abnormalities in carbohydrate tolerance and the post therapy curve was frequently lower than tolerance studies without adrenal steroids

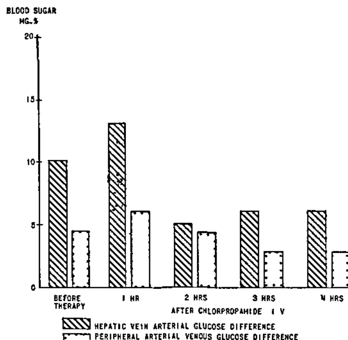


FIGURE 7 Compos te of hepat c vein arterial and per pheral arterial venous glucose d fference in 6 pat ents ithout l ver d sease after adm nistration of 1 0 gm of chlor propam de intravenously

Similar results were obtained with oral and parenteral tests (FIGURES 9 and 10)

endogenously secreted steroids is not the basic mechanism of action of chlor propam de it is evident that in adequate dosage this drug prevents blood sugar abnormalities resulting from short term receipt of glucocorticoids We have also found it effective in control of carbohydrate abnormalities in a patient with latent diabetes who required prolonged adrenal steroids for a skin disorder

Effects on epinephrine hyperglycemia The capacity of sulfonylurea derivatives to increase liver glycogen has suggested their usefulness in treating patients with deficient hepatic glycogen resulting from diabetes or liver disease[15] Unlike healthy subjects in whom there is an increase in blood sugar of 40 mg per cent or more after administration of epinephrine or glucagon, those with liver injury and poor glycogen stores frequently show

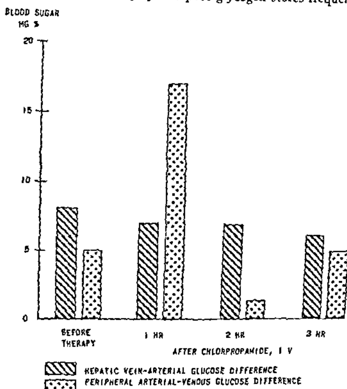


FIGURE 8 Hepatic vein arterial and peripheral arterial venous glucose difference in a patient with cirrhosis and vascular shunts

a diminution in anticipated response[16] In most instances there is concomitant increase in hyperglycemic response with an improvement in biochemical and histological abnormalities of the liver This improvement may be accelerated by glucocorticoids in nondiabetic patients and by insulin in diabetic patients Sulfonylurea derivatives produce a similar effect in diabetes (FIGURE 10) Evaluation of the effect of chlorpropamide in 4 subjects with chronic active liver disease, normal fasting blood sugars, and decreased glycogen stores revealed a significant increase in response to epinephrine not associated with improvement in other biochemical tests in 2 patients

Response of liver disease and diabetes Diabetes and hepatic dysfunction frequently coexist, and assembled evidence indicates that either of these con

... e pon ble for the other[15] The presence of the

BLOOD SUGAR MG %

HOURS FASTING ½ 1 2 3

GLUCOSE TOLERANCE 1 75 GRAM OF GLUCOSE PER KILOGRAM OF BODY WEIGHT

GLUCOSE TOLERANCE WITH 15 MILLIGRAMS PREDNISONE 6 AND 2 HOURS BEFORE INGESTING GLUCOSE

PREDNISONE GLUCOSE TOLERANCE WITH 1 0 GRAM OF CHLORPROPAMIDE AT TIME OF INITIAL PREDNISONE

FIGURE 9 Effect of chlorpropamide on the oral adrenal steroid glucose tolerance test

of diabetics in this category have abnormal liver function tests, abnormal biopsies, or both in our experience Only by long term serial studies is it possible to evaluate hepatic effects of this drug To date, we have not encountered any clinical biochemical or histological changes resulting from chlorpropamide therapy

Many have suggested that subclinical metabolic changes involving the liver are responsible for adult onset diabetes The association of liver injury and diabetes in hemochromatosis pancreatitis, and other metabolic and infectious processes is well documented The diabetic may develop

intercurrent viral hepatitis biliary obstruction or hepatic changes due to heart failure however nutritional deficiency is responsible for most instances of liver disease[17] in this condition The diabetic patient is similar to the insulin maintained depancreatized dog in that lipotropic deficiency produces a fatty liver and may eventually lead to cirrhosis [18] In our experience diet and insulin or sulfonylurea derivatives have been effective in control of both

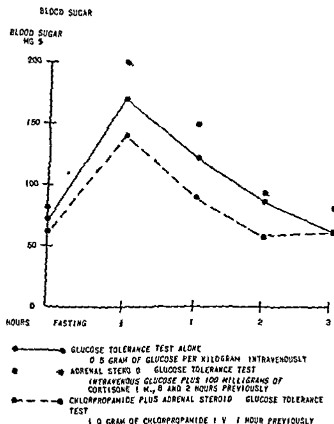


FIGURE 10 Effect of chlorpropamide on the intravenous adrenal steroid glucose tolerance test

the carbohydrate and hepatic abnormalities in most patients with liver dysfunction secondary to poor nutrition

Occasionally liver disease may be responsible for a diabetic syndrome that will disappear with improvement in biochemical and histological abnormalities [19] The prednisone glucose tolerance test performed after the appearance of evidence of diabetes has permitted division of such patients into a group where post therapy adrenal steroids produce a diabetic curve and one where glucocorticoids do not alter glucose tolerance The mechanism of the diabetic syndrome in the latter group is unknown but may be due to a dis

turbance in regulation of blood sugar caused by interference with hepatic production or the use of humoral substances responsible for glycolysis and peripheral utilization of glucose [20, 21]

In our experience patients with this condition readily respond to carbutamide tolbutamide and chlorpropamide Theoretically, sulfonylurea derivatives may be the drugs of choice in such patients because of their ease

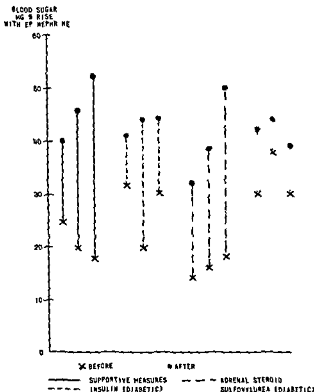


FIGURE 11 Effect of various therapeutic measures on epinephrine induced hyperglycemia in diabetes and liver disease

relationship of encountered hyperglycemia and glycosuria to liver injury This patient was found to have hyperglycemia and glycosuria which disappeared on carbutamide and a diet of 300 gm carbohydrate 100 gm protein and 100 gm fat There was no history of familial diabetes or clinical and laboratory evidence of pancreatitis or endocrine disorder An initial liver biopsy showed marked fatty metamorphosis, which disappeared after

2 months of treatment Post therapy studies demonstrated an increase in epinephrine induced hyperglycemia over control levels a subsequent prednisone glucose tolerance test was normal

Summary and Conclusions

Chlorpropamide blood levels appear to depend primarily on its absorption and urinary excretion and secondarily on tissue storage and metabolism

This drug usually produces an earlier decrease in hepatic venous glucose than arterial glucose with little effect on peripheral arteriovenous glucose difference Selected *patients with cirrhosis* and *vascular shunts may exhibit* a smaller than normal hepatic activity and a significant peripheral effect

Chlorpropamide inhibits glucocorticoid induced hyperglycemia and may

[illegible]

respond to sulfonylurea derivatives makes it difficult to evaluate hepato toxicity of these drugs They have been valuable in management of both the carbohydrate disturbance and hepatic abnormalities seen in diabetic patients with secondary hepatic changes as well as diabetes made overt by liver injury

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CLINICAL EXPERIENCES WITH CHLORPROPAMIDE
A DOUBLE-BLIND STUDY

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Pennsylvania Hospital, Philadelphia, Pa

To determine the clinical effectiveness of chlorpropamide as a hypoglycemic agent, a double blind trial was conducted in the diabetic outpatient clinic at Pennsylvania Hospital After completion of the trial, the patients were asked to continue in a follow up study of the drug The response to chlorpropamide as measured by the eventual state of diabetic control during the follow up period was then compared with the results obtained during the trial and also with the control obtained with previous diabetic therapy The results of this study indicate that chlorpropamide is a potent hypoglycemic agent that produced excellent control of diabetes in most patients

Method

The double blind study was divided into 3 periods of 2 weeks each One week before starting, the patients stopped insulin or tolbutamide if they were taking either of these, but continued their regular diabetic diets The first 2 week period of the trial was a control for all patients while, in the second and third 2 week periods, they received chlorpropamide, 1 0 gm daily, or a placebo The order of receiving chlorpropamide and placebo was chosen at random Using this sequence in the study, it was possible for a patient to have no therapy other than diet for a period of 5 weeks In selecting patients to participate in the trial, we chose only those who, we felt, could [illegible]

The patients came to the clinic in a fasting state at 8 A M and had blood drawn for a blood sugar and appropriate blood chemistries They were then taken to a hospital dining room and were given the same 400 calorie [illegible] were interviewed [illegible]

procedure was repeated for 6 weeks with each patient The patients [illegible] tested their urine before meals and, at bedtime, with Tes Tape,* recorded the results, and brought the tapes to the clinic for our count Chlorpropamide, 1 0 gm daily, and the placebo substance were administered identically to all patients as 2 tablets before breakfast After the trial, patients were seen at 4 week intervals in the clinic while being continued on chlorpropamide During the follow up period, the dose of chlorpropamide was changed as indicated by the state of diabetic control Postprandial blood sugars were not done during the follow up study

All patients on chlorpropamide had additional routine laboratory studies

* Eli Lilly and Company, Indianapolis, Ind

performed These comprised a complete blood count, including hemoglobin, hematocrit, white cell count, and differential count, a routine urinalysis, direct and total serum bilirubin, alkaline phosphatase, thymol turbidity, and cephalin cholesterol flocculation These studies were done prior to beginning the double blind trial, at the end of the trial, and routinely at 1, 2, 5 and 7 months during the follow up period, or more often if clinical indications warranted Blood sugar determinations were made using the Somogyi Nelson technique, while all other studies were done by the routine methods used in the blood chemistry and hematological laboratories of the Pennsylvania Hospital

The results of the double blind study were assessed by averaging the fasting and postprandial blood sugars separately in every patient for each 2 week period These values were then compared for the 3 periods This permitted a comparison of both the fasting and the postprandial blood sugar values between the control and placebo periods, between the control and the chlorpropamide periods and between the placebo and the chlorpropamide periods An adequate response was taken to be a decrease in both the fasting and postprandial blood sugars of 20 per cent or more when levels in the treatment period were compared to those obtained in placebo and in control periods

The final state of control of the diabetes achieved during the follow up period was classified as good, fair, and poor The criteria used were as follows good control, fasting blood sugar under 120 mg per cent and no glycosuria, fair control, fasting blood sugar below 150 mg per cent and minimal glycosuria poor control, all conditions worse than this Those patients who had taken tolbutamide prior to this study also had their diabetic control while on that drug assessed in the same way so that a comparison between the level of control on tolbutamide and on chlorpropamide could be made

Results of Double Blind Trial

Thirty two patients completed the trial The clinical characteristics of the group are given in TABLE 1 All patients were in the older age group,

TABLE 1
CLINICAL CHARACTERISTICS OF PATIENTS IN CHLORPROPAMIDE TRIAL

Age (years)	59 8 av	45 78 range
Age onset of diabetes	52 6 av	32 75 range
Duration of diabetes	6 6 av	0-31 range
Previous therapy		
Insulin	23 patients	10-70 U range
Diet only	4 patients	
Tolbutamide	19 patients	Just prior to trial

with an adult type of diabetes There were 5 men and 27 women Twenty three had taken insulin at some time and 19 were taking tolbutamide immediately prior to the trial

The response to chlorpropamide is shown in TABLE 2 The results are divided into 2 groups 23 patients who received the placebo in the 2-week period before chlorpropamide, and 9 who received chlorpropamide before the placebo Thirteen, or 56 per cent of those in the first group showed a fall in blood sugar of 20 per cent or more when the fasting and postprandial blood sugar values during chlorpropamide administration were compared

TABLE 2
RESPONSE TO CHLORPROPAMIDE

Order of trial	No patients	Adequate response	Inadequate response
Placebo first	23	13 (56%)	10 (44%)
Chlorpropamide first	9	8 (90%)	1 (10%)
Total	32	21 (65%)	11 (35%)

Adequate response = fall in fasting and postprandial blood sugar values of 20 per cent or greater compared to control and placebo levels

TABLE 3
RESPONSE TO CHLORPROPAMIDE PERCENTAGE CHANGE OF BLOOD SUGAR

Order of trial	Placebo Control		Chlorpropamide Control		Chlorpropamide Placebo	
	FBS	PPBS	FBS	PPBS	FBS	PPBS
Placebo first Adequate response	+2%	+2%	−27%	−23%	−28%	−22%
Placebo first Inadequate response	+10%	+16%	+2%	+11%	−10%	+1%
Chlorpropamide first Adequate response	−22%	−19%	−33%	−31%	−11%	−10%

to the values of the placebo and the control periods Eight of the 9 patients who received chlorpropamide first showed an average decrease in blood sugar of more than 20 per cent, but in this group the values obtained on chlorpropamide were compared only with the control values

TABLE 3 shows the average percentage change in blood sugar values in the 2 groups The 3 comparisons are given separately for both fasting and postprandial blood sugar levels When placebo administration followed the control period, the blood sugar values were higher during the placebo period However, when chlorpropamide was given before the placebo, the placebo blood sugar levels were about 20 per cent lower than the control levels This was assumed to be due to the persistence of the hypoglycemic

effect of chlorpropamide after the drug was stopped An adequate response in this group of patients was judged on a comparison of the control and the chlorpropamide periods only It can also be seen that in each group the decrease in blood sugar was slightly greater in the fasting than in the post-prandial levels However, the difference was so slight that, in a patient who has fasting hyperglycemia, determination of the fasting blood sugar alone would have been an adequate indicator of response

Follow-Up Study

Thirty of the 32 patients continued to take chlorpropamide after the trial was completed Of the 2 patients not followed, 1 was hospitalized with a cerebral thrombosis 5 weeks after the trial, the other refused to return TABLE 4 gives the final results to date for the remaining 30 patients Twenty-five of these achieved good control, although the maximum response was

TABLE 4
RESPONSE TO CHLORPROPAMIDE FOLLOW UP OF 30 PATIENTS

Present state of diabetic control	No of patients	Duration until best control in weeks				
		4 or less	6	8	12	16 or more
Good FBS under 120 mg per cent No glycosuria	25	13	3	2	6	1
Fair FBS under 150 mg per cent Slight glycosuria	4	4				
Poor	1					

Of 11 patients with inadequate response during trial, 10 were available for follow up and 9 achieved good control

TABLE 5
RESPONSE TO CHLORPROPAMIDE COMPARISON OF CONTROL WITH TOLBUTAMIDE AND CHLORPROPAMIDE

Tolbutamide control		Chlorpropamide control		
		Good	Fair	Poor
Good	7	7	0	0
Fair	7	7	0	0
Poor	5	4	1	0
Total	19	18	1	0

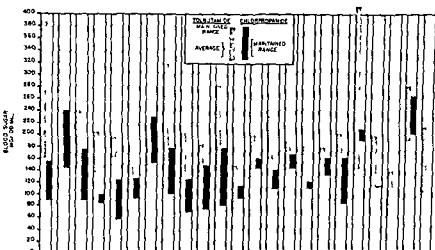


FIGURE 1

R F—46YR COL ♀—DIABETES 6YR

FASTING BLOOD SUGAR MG %

GLYCOSURIA UNITS

WEIGHT POUNDS

INSULIN 40U/DAY | CARBUTAMIDE 1.0GM/DAY | ORINASE 2.0GM/DAY | CONTROL | CHLORPROPAMIDE TRIAL 1.0GM/DAY | CHLORPROPAMIDE 0.5GM/DAY

FIGURE 2

often delayed for several weeks Probably this was due to the fact that the dose of chlorpropamide was usually reduced to 0 5 gm daily after the trial proper was completed The 1 patient who failed to show a good response was a 45-year old Negro female with diabetes of 5 years' duration who had required 50 U of insulin daily for indifferent control before the study

Nineteen patients were taking tolbutamide before the chlorpropamide trial started The effectiveness of diabetic control with these 2 drugs is compared in TABLE 5 Seven patients showed good control with both drugs

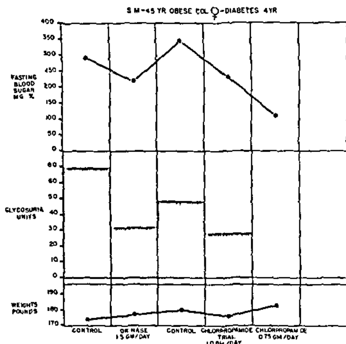


FIGURE 3

Of 12 patients not well controlled on tolbutamide, all showed improvement with chlorpropamide, and with one exception, the final state of diabetic control was very satisfactory FIGURE 1 illustrates this by comparing the range of fasting blood sugar determinations on the two drugs In almost every case the blood sugar values are substantially lower with chlorpropamide There was no correlation between success or failure with tolbutamide and the dose of chlorpropamide necessary to achieve good control FIGURE 2 presents the chronological history of a patient who was treated successively with insulin, carbutamide, tolbutamide, diet alone, and chlorpropamide Average values for fasting blood sugar, glycosuria, and weight are given for each period of therapy The patient showed poor control on 2 0 gm tolbut

amide daily but has been very effectively controlled with a single daily 0 5 gm dose of chlorpropamide FIGURE 3 shows the results of oral therapy in a gluttonous woman who might be controlled by a low calorie diet and weight reduction Neither threats nor bribes nor tolbutamide was effective in controlling her appetite or her diabetes, however on a dose of chlorpropamide that has now been reduced to 0 5 gm daily, she has an average fasting blood sugar of 100 mg per cent and no glycosuria Her appetite is unchanged and she continues to gain weight

Toxicity

The side effects and possible toxic reactions noted with chlorpropamide are listed in TABLE 6 Six patients complained of symptoms suggestive of

TABLE 6
RESPONSE TO CHLORPROPAMIDE SIDE EFFECTS AND TOXIC REACTIONS

Side effect or reaction	No of patients
Hypoglycemic symptoms	8
Fasting blood sugar below 60 mg per cent	4
Hunger	5
Gastrointestinal disturbance	0
Skin rash (transient)	2
Liver function test abnormal (transient)	3
Blood count or urine tests abnormal	0

hypoglycemia and in 4 instances fasting blood sugar values below 60 mg

hazard with chlorpropamide therapy Two patients developed a [illegible] itching maculopapular eruption There was no associated eosinophilia and [illegible] despite continuing

Summary

Thirty two patients participated in a double blind trial of chlorpropamide All had an adult type of diabetes but every patient also had glycosuria and fasting hyperglycemia during the control period of no therapy Twenty-one or 65 per cent of the group had a decrease in both fasting and postprandial [illegible] Thirty patients of the 5 gm

or less daily given in a single dose The maximum response to chlorpropamide may be delayed until 4 weeks or more after therapy is started Comparison of diabetic control between tolbutamide and chlorpropamide in the same patients shows that good control often can be achieved with chlorpropamide when tolbutamide has been proved ineffective Chlorpropamide was a more potent hypoglycemic agent than tolbutamide Toxic reactions were seen in 5 patients but in every case the manifestation of toxicity subsided without stopping therapy Hypoglycemic reactions occasionally severe may be provoked by chlorpropamide and the dosage must be individually adjusted

Acknowledgments

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LONG TERM THERAPY WITH CHLORPROPAMIDE

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The influence of chlorpropamide has been evaluated in 56 diabetic patients with reference to changes in hyperglycemia, glycosuria, liver function, the hematopoietic system, and general health

Materials and Methods

The subjects were selected largely from the diabetic clinic of St Boniface Hospital As a group they tended to have little respect for dietary restrictions Before inclusion in the study an attempt was made to categorize the diabetes as stable (adult, obese, nonketotic, and senile) or labile (juvenile, thin, ketotic, and brittle) Using this clinical assessment, we felt that 49 subjects had stable diabetes

The initial response to chlorpropamide was assessed in hospital on 37 patients The other 19 were studied as outpatients The laboratory tests done prior to therapy and at intervals thereafter were a complete urinalysis, hemogram oral glucose tolerance (Folin Wu) blood urea nitrogen, plasma prothrombin time, serum alkaline phosphatase (King Armstrong), glutamic oxaloacetic acid transaminase, phosphohexose isomerase thymol turbidity and flocculation, cephalin cholesterol flocculation, cholesterol, and esters and Bromsulphalein excretion The radioactive iodine uptake by the thyroid was measured in 5 of these subjects

Depending on the type and severity of the diabetes, chlorpropamide administration was begun in one of several ways the drug was given and the insulin gradually reduced insulin was discontinued 24 hours before giving chlorpropamide, insulin was discontinued for several days, patients who had not been using insulin were given the drug after a preliminary assessment on a constant diet In all cases 1 0 gm chlorpropamide was – on the first day and 0 5 gm daily until the hypoglycemic effect could be

Results and Discussion

Thirty nine of the 56 subjects have received chlorpropamide for at least 4 months, which is our criterion for long term therapy The 7 patients adjudged to have labile diabetes all failed to show a hypoglycemic effect with chlorpropamide either alone or in combination with insulin They were given the drug from a minimum of 4 days to a maximum of 20 days We concluded that chlorpropamide, like the other sulfonylurea drugs, does not control the labile diabetic However, our dose schedule for initiating therapy with chlorpropamide was not varied, and it is possible that a higher initial

TABLE I
ANALYSIS OF PATIENTS WITH STABLE DIABETES

Age and sex	Diabetes		Insulin			Chlorpropamide			
	Duration (years)	Complications	Duration (years)	Daily dose (units)	Control*	Duration (days)	Dose (gm.)	Control*	Comments
56M	New	+	0	0		? 20†	0 5	G‡	Sudden death M infarct
74M	2	+	Only with acute cholecystitis			10	0 5	G‡	Recurrent cholecystitis
66M	1½		0	0	P	7	0 5	?‡	Obstructive jaundice due to Ca pancreas
70M	12	+	0	0		20	0 75	G‡	
46F	4		0	0	P	40	0 5	?‡	P on 0 5 gm
47F	6	+	0	0	P	45	0 75	?‡	P on 0 75 gm
56M	9	++	0	0	P	7	2 0	P	
68F	14		14	75	P	21	2 0	P	
65M	16	+	12	40	P	10–16†	2 0	P	Diarrhea first course none later
66F	10	−	10	30	C	14	0 75	P	Anorexia drowsy ? reaction
66F	5	+	5	70	P	120	1 5	P	Hypoglycemic effect in hospital
45F	9	−	4	35	P	200	2 0	G	Initial success operation
42F	3		3	30–40	F	270	1 0	G P	Initial success
72F	10	+++	5	25	P	230	1 0	G	Died post op cholecystectomy
41M	1½	−	1½	35	G	90	0 5	G	Triamcinolone 100 mg./day died leukemia
59F	19	+	0	0	F	300	0 5	C	
81F	2	+	½	15	F	180	0 25	C	
71F	7	+	5	30	P	180	0 75	F	Gangrenous toe
63F	23	++	15	30	F	180	0 5	G	Hyperthyroidism
76F	2	++	1½	15	G	120	0 5	G	Cellulitis
66F	3		3	50	P	150	0 5	G	
72M	5	++	0	0	F	270	0 25	G	Transurethral resection
63F	9	+	1§	25	P	240	0 75	C	Pneumonia
75F	22		18	30	G	270	0 25	C	
71F	5	+	5	30	P	240	1 0	G	
52M	2	−	0	0	P	2 0	0 5	F	Electroshock therapy
74M	6	+	0	0	P	270	0 75	C	
49F	5	−	2	30	P	150	1 0	P	
65M	1	+	0	0	P	180	0 5	C	
68F	8	++	4	20	P	270	0 25	G	
79M	5	+	4	30	P	240	0 5	G	
50F	7	+	5	50	P	270	0 75	F	Myocardial infarct Abdominal burn
62F	5	+	¾	30	P	240	0 5	G	Cerebrovascular accident
49F	1	−	0	0	P	180	0 25	G	
77M	?Recent	½	½	35	F	180	0 5	F	
71F	5	+	5	35	F	135	0 25	G	
70F	16	++	0	0	F	120	0 25	G	
67F	1	−	½	25	F	300	0 25	G	
55F	5	++	5	10–30	F	180	0 25	G	
74F	12	++	0	0	P	130	0 75	G	
50F	?	−	2	15	P	240	0 5	G	
63F	2	++	½	35	G	200	0 25	C	
70F	7	+	6	80	?	180	1 5	F	Carcinoma colon resected
67F	?	−	4	20	P	240	0 5	G	
64F	8	+	2½	35	?	210	0 75	G	
73F	3	+	0	0	P	190	0 25	G	
60F		+	½	13	F	270	0 25	G	
64F	6	−	0	0	P	210	0 25	G	
65F	New		0	0		120	0 5	C	

* Control P = poor F = fair G = good
† Chlorpropamide given in 2 courses
‡ Lost to follow up
§ At onset of diabetes

dose might have produced hypoglycemia. No side effects other than ketosis were observed. One labile patient, a 39 year-old female, died 10 days after chlorpropamide was discontinued. The cause of death was a pulmonary embolus following electroshock therapy. The consulting psychiatrist felt that her psychosis was not drug induced.

Some of the pertinent data on the 49 stable diabetics are shown in TABLE 1. The complications indicated as +++ refer to retinopathy, neuropathy, nephropathy and other occlusive vascular disease. By good control we mean

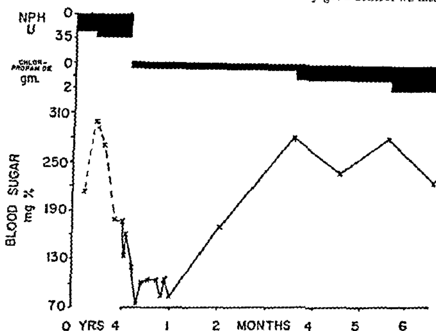


FIGURE 1. Patient H. M., female, aged 45, diabetic for 9 years.

aglycosuria and fasting normoglycemia, and by fair control we mean fasting blood sugars (Folin Wu) between 120 and 160 mg. per cent. Fasting blood sugars over 160 mg. per cent indicate poor control. The patients varied in age from 41 to 81 years, with a mean age of 63. Ten of these subjects did not satisfy our criterion for long term therapy. Six were lost to follow up, 3 failed to show a hypoglycemic effect on 2.0 gm. chlorpropamide daily, and 1, who remained hyperglycemic on 0.75 gm. daily, became anorectic and drowsy. Of those lost to follow up, 1 died suddenly of a myocardial infarct diagnosed post mortem. His initial response had been good and he may have been hypoglycemic at the time of infarction. One died of *E. coli* septicemia during his third bout of acute cholangitis. He was normoglycemic on 0.5 gm. chlorpropamide between the second and third attacks. The drug was discontinued at the onset of symptoms 2 weeks prior to death. At post mortem, multiple liver abscesses were found, together with common duct calculi and a contracted gall bladder. A third patient developed

1 showed good initial control on 0 5 gm and 2 showed poor initial control on 0 5 gm

All of the 39 long term subjects showed an initial hypoglycemic effect Three were late failures FIGURE 1 shows the decrease in fasting blood sugar level in a 45 year old obese female who was changed from insulin to

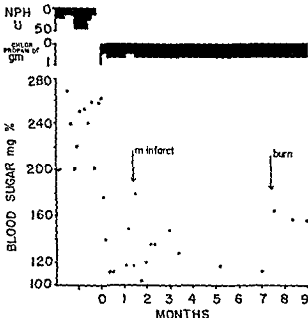


FIGURE 2 Patient D M female aged 50 diabetic for 7 years

pital but control at home was better than on previous therapy The other 30 patients remained well controlled at home

Two of the long term subjects have died one of leukemia and the other of subphrenic abscess following cholecystectomy for chronic recurrent cholecystitis Both patients showed good control on chlorpropamide Good control was maintained on the leukemic patient during a 3 week period when he received triamcinolone (100 mg /day)

In the long term group 9 further medical events have occurred 1 myo

shows the blood sugar response in a 50 year old female who had first a myocardial infarct and later a second degree scald of the abdomen The possible role of chlorpropamide in the production of these fatal and nonfatal events is not known, but no direct correlation could be detected

Oral glucose tolerance tests done 24 hours following therapy showed a hypoglycemic effect, but no other difference in the type of curve FIGURE 3 shows 1 pretreatment and 3 treatment curves in a 52 year-old male The subject was on an unrestricted diet during the 7 months between the third and fourth tests The fourth curve shows less hypoglycemic effect

Liver function showed no deterioration except in 3 patients who died In those 3 a satisfactory explanation of the pathology was found (see above)

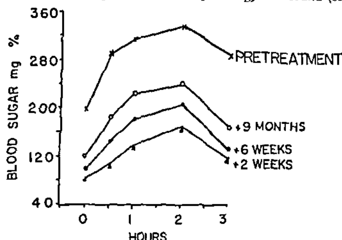


FIGURE 3 The effect of chlorpropamide on oral glucose tolerance Patient A K male, aged 52 diabetic for 2 years

In 3 other patients Bromsulphalein excretion improved during drug administration Hematological studies showed no change in hemoglobin and no evidence of agranulocytosis or thrombocytopenia Two patients showed a transient unexplained eosinophilia Chlorpropamide did not change the uptake of radioactive iodine by the thyroid in the 5 patients studied, including the one with hyperthyroidism

Symptoms that may have been due to the drug were diarrhea in one patient and pruritus in another In both cases the drug was continued, with disappearance of the complaint One patient complained of anorexia and drowsiness with hyperglycemia but no acidosis, and the drug was discontinued

An attempt was made to find a correlation between duration of insulin therapy and blood sugar response to chlorpropamide in the stable group Of 18 who had never taken insulin, 10 showed a good response, 1 a fair response, and 1 a poor response Six were lost to follow up Of 18 on insulin less than 5 years, 14 showed a good response, 2 a fair response, and 2 were late failures Of 13 on insulin for 5 years or more, 6 showed good control, 3 showed fair control, and 4 showed poor control Of the 31 who

had used insulin, 5 had taken more than 40 units daily Three of these patients showed improved control with chlorpropamide Ten patients in the stable group had previously used tolbutamide Of these, only 3 had improved on tolbutamide, whereas 7 improved on chlorpropamide This

13 used 0 5 gm , 6 used 0 75 gm , 3 used 1 0 gm , and 1 used 1 5 gm daily Final adjustments in dosage have not yet been made

Summary

We could neither control the hyperglycemia nor prevent ketosis with chlor propamide in 7 labile diabetics Thirty six of 43 stable diabetics were controlled better with chlorpropamide than with previous therapy Although the mortality and morbidity rates were high, chlorpropamide could not be implicated No deterioration of liver, hematopoietic, or thyroid function could be determined Diarrhea, pruritus, and anorexia each occurred in one patient Neither the amount nor the duration of insulin therapy correlated with the hypoglycemic response to chlorpropamide Some tolbutamide fail ures were controlled by chlorpropamide The maintenance dosage of chlor propamide averaged 0 5 gm daily

Acknowledgments

We acknowledge with pleasure the help of M C Blanchaer and Paul Green, staff members, St Boniface Hospital, Winnipeg, Canada

CLINICAL EVALUATION OF CHLORPROPAMIDE IN DIABETES MELLITUS

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The use of the sulfonylureas for the treatment of diabetes mellitus is now well accepted Numerous studies dealing with the indications, contraindications, and clinical results of oral treatment with carbutamide and tolbutamide have been published Therefore, in a clinical study of a new drug chlorpropamide, 1 propyl 3-(*p* chlorobenzenesulfonyl)urea, it seems unnecessary to survey the literature, even for the sake of comparison, since the fundamental therapeutic data are so well known

Materials

Chlorpropamide was administered to 80 diabetics (38 males, 42 females) The ages of the patients, dates of onset, duration of diabetes, nutritional

Table 1

Age and Nutritional Status in 80 Patients Treated with Chlorpropamide

Nutritional status	Age in years			Total	Percentage
	<20	21–40	>40		
Obese	—	4	44	48	60 0
Normal	—	5	9	14	17 5
Underweight	2	5	11	18	22 5
Total	2	14	64	80	100 0

Table 2

Age of Onset and Years of Diabetes in 80 Patients Treated with Chlorpropamide

Age of onset	Years of diabetes					Total	Percentage
	<1	1 5	6–10	11 15	16–20		
<20	—	2	—	—	—	2	2 5
21–40	3	8	8	—	4	23	28 7
>40	6	19	12	15	3	55	68 7
Total	9	29	20	15	7	80	100 0

TABLE 3
COMPLICATIONS OF DIABETES AND ASSOCIATED DIAGNOSES IN 80 PATIENTS TREATED WITH CHLORPROPAMIDE

Complications or associated diagnoses	No of cases
Retinopathy	29
Nephropathy	17
Atherosclerosis	42
Blood hypertension	15
Tuberculosis	9
Other diagnoses	10
Patients with 1 or more complications or associated diagnoses	53
Patients without complications or associated diagnoses	27

TABLE 4
THERAPY PRIOR TO CHLORPROPAMIDE IN 80 PATIENTS

Prior therapy	No of cases	
Insulin treatment		61
Insulin only	35	
Insulin and oral treatment (nonsimultaneous)	26	
Oral treatment only		8
New cases without treatment		4
Old cases without treatment		7
Total		80

TABLE 5
YEARS OF INSULIN AND DOSE IN 61 PATIENTS TREATED WITH CHLORPROPAMIDE

Years of insulin	Dose of insulin (units)			Total
	<20	21–40	>41	
<1	3	11	4	18
1–5	4	15	1	20
6–10	—	8	1	9
11–20	1	5	1	7
Not determined	3	4	—	7
Total	11	43	7	61

condition complications and associated maladies, and previous treatment are illustrated in TABLES 1, 2, 3, 4, and 5

METHOD

Thirty four cases were first treated while hospitalized, all the other cases were outpatients The diet, adjusted to the theoretical caloric requirements, varied between 150 gm and 250 gm of carbohydrates The initial dosage of chlorpropamide was 2 gm (23 cases) and 1 5 gm (5 cases) daily, based on the usual dosage schedules for the other sulfonylureas As our evaluation continued we found that we could reduce the initial dosage to 1 gm (49 cases) and 0 5 gm (3 cases) daily In general, divided doses were employed, except during the maintenance period, when the dose was reduced to 0 25 gm daily In the case of 2 patients, both chlorpropamide and insulin were given

The outpatients were examined weekly at first, and then monthly In these cases the blood sugar was determined during a fasting state and urine sugar was determined from a 24 hour pooled sample (glycemia determined by the Solomos method) Before and during the course of treatment tests were made of hepatic, hematopoietic, and renal functions (flocculation, alkaline phosphatase prothrombin, Bromsulphalein retention, hemogram, and complete urine tests)

The first dose was administered as a single dose and the hypoglycemic action of the drug was determined in 76 cases at 3, 4, 5, and 24 hours (selection test) On the basis of these results the potency of the drug was established The patients fasted during the first 5 hours of study In addition to observing immediate action, we wished to determine whether the results of this test would relate to the results of previous treatment Serum chlorpropamide also was tested during the course of the treatment *

RESULTS

Hypoglycemic Action of the Drug

Selection test Response as observed in the selection test was based on blood sugar determinations on samples taken 5 hours after the initial dose was administered (final glycemia) On the basis of our prior experience with other sulfonylureas,[1] the result was classified as good when the final blood sugar was lower than 1 60 gm per cent or a drop of more than 50 per cent from the initial figure occurred The result was considered fair when the blood sugar was between 1 61 and 2 gm per cent or when a drop of between 25 and 50 per cent from the initial figure occurred, the result was termed poor when the final blood sugar was over 2 gm per cent or when a decrease of less than 25 per cent from the initial figure occurred

A significant hypoglycemic effect appeared in 52 patients (68 4 per cent), the other 24 did not respond to the drug FIGURE 1 presents the curves of the average values obtained in the patients who responded to and those who did not respond to the drug The tests carried out with 1 and 2 gm of chlorpropamide are shown separately The greatest decrease in blood sugar was

* Using the technique communicated to us by Chas Pfizer & Co Inc , Brooklyn, N Y

the other sulfonylureas chlorpropamide has an immediate hypoglycemic action

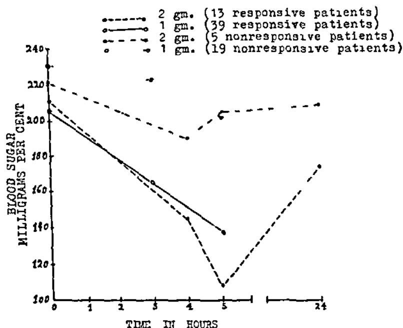


FIGURE 1 Average blood sugar response to different doses of chlorpropamide

TABLE 6 presents the results of four selection tests conducted on patients who received chlorpropamide in which serum levels of the drug were also determined at the same time

It may be seen that in the 3 cases with good hypoglycemic response there were considerable quantities of chlorpropamide in the blood during the early hours of the test These values increased even more at 5 hours The increase is more rapid in some cases There seems to be a period of latency of action since the blood sugar values rapidly fall at 5 hours after 0 5 or 1 gm are administered even though in the serum at the 3 hour determinations there are greater quantities of chlorpropamide than are usually observed in patients who have responded to the drug and are receiving maintenance dosage The case with poor response (case 4) showed serum chlorpropamide levels at 3 and at 24 hours that were higher than those obtained in other patients who had a favorable response In addition to the usual differences

TABLE 6
CHLORPROPAMIDE SERUM LEVEL AND BLOOD SUGAR RESPONSE IN 4 PATIENTS, AFTER ADMINISTRATION OF A SINGLE DOSE OF 1 0 GM

	0 Hour blood sugar	3 Hours		5 Hours		24 Hours	
		Blood sugar	Serum chlorpropamide mg %	Blood sugar	Serum chlorpropamide mg %	Blood sugar	Serum chlorpropamide mg %
Case 1	2 22	1 66	4 03	1 14	8 21	1 25	6 59
Case 2	1 66	1 53	3 90	1 43	5 30	1 90	9 89
Case 3	3 08	2 00	6 40	1 29	9 23	Not done	Not done
Case 4	2 10	2 67	7 41	1 90	Not done	2 85	6 78

Case 1 Record 492 (E R) Case 2 Record 509 (C Z)
Case 3 Record 501 (E M) Case 4 Record 381 (P V)

TABLE 7
TIME OF CHLORPROPAMIDE TREATMENT REQUIRED TO OBTAIN CONTROL OF BLOOD SUGAR OR GLYCOSURIA*

Time of treatment	Blood sugar			Percentage	Glycosuria			Percentage
	No cases		Total		No cases		Total	
	2 gm	1 gm			2 gm	1 gm		
1st Week	9	23	32	58	9	22	31	58 4
2nd Week	5	12	17	31	2	11	13	24 5
3rd Week	2	2	4	7 3	—	1	1	1 9
4th Week	—	—	—	—	1	3	4	7 5
5th Week	1	1	2	3 6	1	3	4	7 5
Total	55 Cases			99 9	53 Cases			99 8

* Blood sugar below 150 mg per cent no sugar in 24 hour urine specimen

in response that can be expected due to the different degrees of diabetes, there also may be variations caused by differing rates of absorption and excretion

Normalization of blood sugar and disappearance of sugar from the urine Blood sugars were returned to normal levels in 55 of the 80 patients Of these, 58 per cent occurred in the first week (16 per cent in the first 4 days), and 31 per cent in the second week Disappearance of sugar from the urine occurred at similar times (TABLE 7) There was no evident difference

between those cases receiving 2 gm or those receiving 1 gm during the initial treatment period Compensation was somewhat more prompt in the hospitalized cases than in the outpatients, possibly because of better observance of the diet

Clinical Results

Results are classified as good, fair, and poor This is the same standard as is used in other publications of this department, with the omission of the category excellent, no curves having been taken for postprandial blood sugar during the day (TABLE 8)

TABLE 8
STANDARD OF DIABETIC CONTROL*

Control	Fasting blood sugar mg per cent	24 Hours glycosuria gm	2 Hours postprandial blood sugar mg per cent
Excellent	<150	0	<180
Good	<150	0	<225
Fair	150–200	<20	<250
Poor	>200	>20	>250

* 180 to 200 gm of carbohydrates in the diet

TABLE 9
RESULTS OF CHLORPROPAMIDE TREATMENT IN 80 PATIENTS

Results	No Cases	Percentage
Good	44	55 00
Fair	23	28 75
Poor	13	16 25
Total	80	100 00

The results have been analyzed in relation to several factors the age of the patients age of onset of the disease, duration of the disease the patient's nutritive state, complications and associated disorders previous treatment, especially with insulin, introductory and maintenance doses of the drug and the serum level of the drug The results of the selection tests have been compared also with the results of the treatment

The analysis has not taken into account the differences in reaction between youthful diabetics and middle-aged obese diabetics, these differences are well known, but in practice it is difficult to classify many patients according to rigid categories without doing violence to the facts of the case In the present analysis greater emphasis has been given to the nutritional state

of the patient a fact that can be determined readily Results of treatment are presented in TABLE 9

In practice according to the criterion adopted patients in whom we observed either a good or a fair response were considered subjects for long term treatment with this reservation the fair cases must be subject to rigid observation since it is possible for such cases to develop inte[illegible] es as time passes even when such development [illegible] for example as dietary d[illegible] hat appr[illegible] ith

Age as Related to Response

Treatment failed in the 2 patients under 20 years of age In the 21 to 40 year group and the group over 40 the percentages of good cases are equal

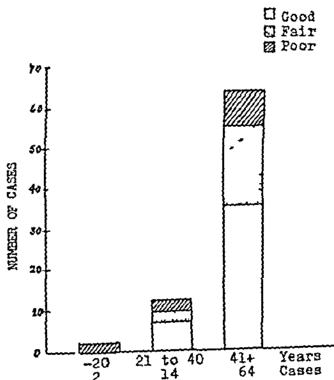


FIGURE 2 Age and results of chlorpropamide treatment in 80 patients.

but the failures are more frequent in the younger patients (21 4 and 12 5 per cent respectively) This tendency reveals a *definite* correlation of age to response but one that probably would have been more striking had the number of patients involved been greater (FIGURE 2)

Onset of Diabetes as Related to Result

This is a complex factor Aside from considering the special characteristic of diabetes this factor includes an unknown the duration of the disease Nevertheless both factors suggest the same conclusions that diabetes in younger patients is habitually refractory to oral treatment and on the other hand that there is a possibility that the disease existed for a longer period of time in those who develop diabetes while young (among our patients were 23 more than 40 years old in whom onset of the disease had occurred between their twenty first and fortieth years) It was observed that when the onset of diabetes was between 21 and 40 years of age the percentage of good results was 47 8 per cent in those in whom the onset of the disease occurred after the fortieth year good results were obtained in 60 per cent of the patients

Duration of the Disease as Related to Response

The groups are rather small for a rigorous statistical analysis but the influence of duration of diabetes seems to be considerable in patients treated with other hypoglycemic drugs

Of those exhibiting a good result 89 per cent were patients whose diabetes was of less than one year s duration Of 22 patients who had had diabetes for more than 10 years 12 responded well to chlorpropamide while the remaining 10 showed a fair response This result may be due to the interference of factors other than the duration of the disease It may be probable that duration of the disease has little or no direct effect on the result obtained with treatment It may be of importance in long standing cases in an indirect manner such as its influence on modifications of the nutritional state that frequently deteriorates in patients not receiving constant treatment

Effect of Previous Treatment on Response

Of the 11 cases that had never been treated with insulin or other measures [illegible] of the 13 failures had received insulin previously and 6 of them also had received treatment with sulfonylureas However this result should not be regarded as a direct consequence of the previous treatment factor exclusively since 90 per cent of the thin underweight patients had required insulin previously but only 70 per cent of the obese patients had received it and the influence that the nutritional state constitution and type of diabetes exert on the response to oral therapy is well known

FIGURE 4 shows that the duration of insulin therapy constitutes an adverse factor on the response to treatment with chlorpropamide In fact 55 per cent of the patients who had used insulin for less than 5 years had good results with chlorpropamide and only 25 per cent of the patients who had used insulin for more than 5 years had such results It should be noted that in the latter group more than half the patients initially had a fair response

In regard to insulin dosage even though the patients who receive high

dosage are few we found that in the 7 patients studied who had received more than 40 U daily there was no case in which good results were observed with chlorpropamide therapy We obtained 4 fair and 3 poor responses in spite of the fact that 4 patients were obese 2 of normal weight and only 1 was thin This would confirm the fact that high insulin dosage is an adverse factor The number of cases studied in our experiments is too small to permit an analysis of the combined effect resulting from the size of the

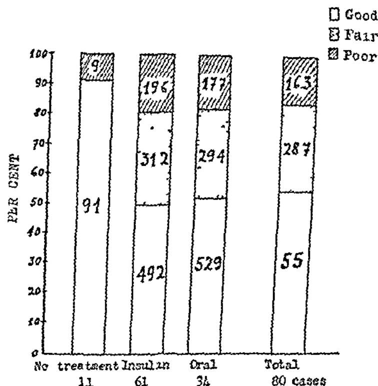


FIGURE 3 Prior therapy and results of chlorpropamide treatment in 80 patients

insulin dosage received and the number of years insulin had been administered

It is interesting to compare results of other types of oral therapy with the results obtained with chlorpropamide TABLE 10 presents the results obtained using chlorpropamide and other orally applied agents (carbutamide and tolbutamide) on 29 patients It may be observed that chlorpropamide brought about compensation in 9 of the 14 cases that were failures under the other drugs and achieved good results in 3 of the 6 cases that were merely fair under the other drugs This is a fact of great therapeutic importance since it offers the possibility of treating patients who have failed to respond to other forms of oral therapy In TABLE 11 it may be seen that the over all

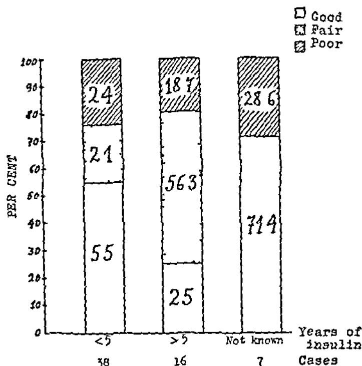


FIGURE 4 Years of insulin and results of chlorpropamide treatment in 61 patients

TABLE 10
COMPARISON OF CHLORPROPAMIDE TREATMENT WITH OTHER HYPOGLYCEMIC AGENTS IN 29 CASES

Chlorpropamide	Other hypoglycemic agents			Chlorpropamide totals
	Good	Fair	Poor	
Good	6	3	5	14
Fair	1	2	4	7
Poor	—	1	5	6
Total	9	6	14	29

TABLE 11
RESULTS OF TREATMENT WITH 3 HYPOGLYCEMIC DRUGS

	Carbutamide		Tolbutamide		Chlorpropamide	
	No cases	Percentage	No cases	Percentage	No cases	Percentage
Success*	169	84	56	72 8	67	83 7
Failure	32	16	21	27 2	13	16 2
Total	201	100	77	100 0	80	100 0

* Including good and fair results

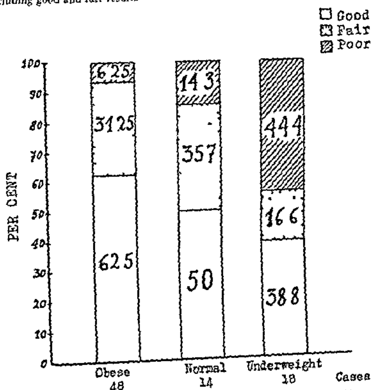


FIGURE 5 Results of chlorpropamide treatment and nutritive status in 80 patients

experience of the authors is that chlorpropamide has demonstrated itself to be as effective as or more effective than carbutamide and tolbutamide in the control of diabetes

Effect of Nutritional Condition on Response

From FIGURE 5 it is obvious that the highest percentage of success (62 5 per cent) was obtained in obese patients just as is the case with the other sul

fonylureas Even in the group between 20 and 40 years of age the percent ... h n the scope of th s

As a matter of fact in ost of the pa ... impossible since they were generally already receiving a moderate low calory diet to conform with their activity and they still required medication to control their blood sugar levels

Effect of Complications or Intercurrent Disorders on Response

In general the presence of retinopathy nephropathia and tuberculosis or other conditions did not affect the response to an appreciable extent as compared with patients who had no pathology other than diabetes mellitus ...

that 7 of the 9 tuberculous patients benefited from chlorpropamide

Effect of Dosage on Response

There was no apparent difference in response to an initial dosage of either 2 gm or 1 gm the latter dose obviated in part the appearance of secondary

TABLE 12
MAINTENANCE DOSE AND RESULTS OF CHLORPROPAMIDE TREATMENT IN 67 PATIENTS

Result	Maintenance dose in mg					Total
	125	250	500	1000	1500	
Good	2	12	15	13	2	44
Fair	—		3	16	4	23
Total	2	12	18	29	6	67
Percentage of Cases	3	18	27	43	9	

reactions especially of a gastrointestinal digestive nature TABLE 12 indicates the maintenance doses employed grouping the patients according to the results It should be noted that among the 13 failures high doses (2 gm daily) were used in 7 cases 1 5 gm doses in 5 cases and 1 gm in only 1 case These were considered failures only after 10 or more days of treatment with these dosages thus ensuring that therapeutic blood levels of the drug had been reached Nevertheless despite the high dosages that often produced intolerance as with the other sulfonylureas poor responses were obtained only *in those patients who were not susceptible to oral therapy for various* reasons

Response as Related to Observation Time

Of the 80 patients studied, treatment was continued in 67 (the 13 failures having been excluded) Of the 67, 62 underwent 30 or more days of treatment

We record a fact known from treatment with the other sulfonylureas in some cases, the results change from good to fair or poor as time passes generally within several months Such patients represent the so called secondary failures If one eliminates the cases in which decompensation is caused by persistent violations of the prescribed diet or by *intercurrent* disease, one is invariably left with cases for the course of which an explanation is difficult With chlorpropamide in 15 patients whose treatment was continued from 90 to 210 days, we did not observe this secondary failure phenomenon, however our observation period is still relatively short

Response as Related to Serum Level of the Drug

From TABLE 13 it may be seen that the serum level of the drug varied from 6 1 to 12 mg per cent in 26 of 34 cases that responded to the drug

TABLE 13
SERUM LEVELS OF CHLORPROPAMIDE IN PATIENTS RESPONSIVE TO THIS DRUG
DETERMINATIONS IN 34 PATIENTS

Serum levels of chlorpropamide	Good			Fair			Total
	250	500	1000	500	1000	1500	
3 1 6	—	2	—	—	1	—	4
6 1–9	1	5	3	2	1	1	13
9 1–12	1	2	2	—	7	1	13
12 1–15	—	—	2	—	—	1	3
21 1–24	—	—	—	1	—	—	1
Total	2	9	8	3	9	3	34

without any apparent indication that this serum level was related to the response to treatment Two cases receiving 250 mg of chlorpropamide daily had blood levels that did not differ from those obtained with higher dosage so that this small maintenance dose appeared to be as effective as the higher dosage

Comparison of Clinical Results with Earlier Selection Tests

Response to treatment with sulfonylureas may be predicted to a degree, by studying the hypoglycemic response obtained with a single dose of the

It can be seen from TABLE 14 that of the 40 selection tests in which good results were obtained, only 3 had a poor response to chlorpropamide However, it was also observed that in 21 patients who showed a poor response to the selection test, only 5 actually showed a poor response to treatment, which renders the test somewhat valueless when it is compared with the clinical indication

Of greater interest is the test with regard to thin, malnourished patients It may be observed that in this group the result of the test coincides very

TABLE 14
SELECTION TEST, NUTRITIVE STATUS, AND RESULTS OF CHLORPROPAMIDE TREATMENT IN 67 PATIENTS

Selection test	Obese			Normal			Underweight			Total
	Good	Fair	Poor	Good	Fair	Poor	Good	Fair	Poor	
Good	15	6	2	4	3	1	7	2	—	40
Fair	2	2	—	—	—	—	—	1	1	6
Poor	7	7	1	—	1	—	—	1	4	21
Total	42			9			16			67

well with the response to treatment The 9 cases with good tests all were susceptible to treatment, of the 5 tests with poor results, in only 1 case was the result of the treatment fair, the other 4 cases being failures Our experience with thin patients indicates that the test is one that provides a rapid means of selecting those patients susceptible to oral treatment with chlorpropamide

Side Reactions

Side reactions were observed in 24 patients One third of these patients presented 1 symptom only, one third presented 2 or 3 symptoms simultaneously and the last third, 4 or more symptoms

in

di-

15 cases (headache, asthenia, drowsiness, dizziness or vertigo, and muscular pains) The most frequent symptoms were epigastric distress (16 cases) and asthenia (14 cases) The frequency of the other symptoms varied between 2 and 7 cases

Cutaneous manifestations were mild In 1 case they disappeared spon-

In the 15 patients who exhibited general symptoms, these were mild in 60 per cent, moderate in 15 per cent, and intense in 25 per cent of the cases

Side reactions usually appeared between the third and fifth days of treatment (range 1 to 11 days), and lasted 4 to 5 days (range 2 to 14 days) They generally appeared when the patient had received a total of between 5 and 10 gm of the drug (range 1 to 16)

With daily doses of 2 gm we observed reactions in 38 4 per cent of the cases, with 1 gm, in 21 per cent With a dose of one half gram only, we observed 1 case of pruritus (which disappeared spontaneously) and no other reaction The reactions were twice as frequent in the patients who responded poorly to treatment This is understandable, since these patients received much higher doses for a longer period of time in an attempt to control the diabetes Six of the 13 failures accounted for 31 per cent of the secondary reactions observed

Since more than 70 per cent of the side reactions were mild or moderate, and since two thirds of those affected presented 1 to 3 symptoms, it was not *necessary to suspend treatment in the majority of the cases* The symptoms disappeared spontaneously or, when the dosage was reduced, in 25 per cent of the cases in only 6 cases was a temporary suspension of treatment necessary, usually for only 2 to 3 days The treatment was discontinued completely in only 2 cases and, in both of these patients, intolerance to the drug coincided with therapeutic failure To some patients we administered the drug in wafer form, which suggested (to the patient) a change of medication This was followed by the rapid disappearance of epigastric distress in patients previously reporting this symptom This occurrence suggested to us the presence of a psychological component that may have been responsible for the occurrence, intensity, and prolongation of side reactions, especially in neuro
pat

du
to
It takes the form of a
palpitations and, more
violent and prolonged
to tetraethylthiuram
bisulfide (Antabuse), and can be interpreted as a consequence of blocking the oxidation of alcohol in the acetaldehyde phase

It is therefore necessary to prevent the patient from consuming any alcohol, even in food We should note at this point that there were patients who
of alcohol (wine) at meals without sustaining

recur, so they continued to use a small quantity of wine

One female patient complained of hypoglycemic symptoms, without laboratory confirmation since they occurred while the patient was at home On two occasions *we noted moderate hypoglycemia* in other patients One of these was a young tuberculous patient, thin, with a diet providing 250 gm of

carbohydrates He began treatment in intense decompensation which has been controlled exclusively by dietary means

Finally, one patient without alcoholic, hepatic, or biliary antecedents showed icterus of the cholangiolithic type after 18 days of treatment Since this case is of especial interest, we reproduce the case history

Patient aged 44, male diabetic since 1952, controlled in a very irregular way with protamine zinc insulin and diet no vesicular or hepatic antecedents On April 1 1958 treatment was initiated with 2 gm daily of chlorpropamide

TABLE 15
TESTS

Tests	3 27 58	4/19 58	4/23 58	5 2/58	5/22/58
Chlorpropamide dose	(2 gm daily 4 1 58 reduced to 250 mg twice daily 4/11/58 continued to 4/18 58)				
Bilirubin direct 1 min	0 3	1 8	1 8	0 2	0 3
Total	0 5	3 3	3 4	0 5	1 0
Total cholesterol	208	no	434	220	—
Alkaline phosphatase	3 2	10 7	10 7	10 7	3 2
(Hanger) flocculation test	(−)	(−)	(−)	(−)	(−)
Prothrombinemia	100%	no	100%	no	—
Bromsulphalein test*	7 6%	no	no	no	0%
Mucoproteins	no	no	4 6	no	no

* Normal retention less than 6 per cent at 45 min

pain without radiation and with no correlation to ingestion of food al o anorexia dark urine and discolored stool were noted On April 15 1958 conjunctival and cutaneous jaundice appeared associated with moderate pruritus Prior to treatment with chlorpropamide the patient had shown normal results in the following tests urine examination hemogram sedimentation test bilirubin determination, alkaline phosphatase test flocculation test Bromsulphalein determination *creatinine clearance and phenol* sulfonphthalein test He was rehospitalized on April 18 1958, and chlorpropamide was discontinued

On physical examination the patient was conscious and lucid pulse 70, regular arterial pressure 110 70, no fever good general condition moderate icterus of skin and mucosa hepatomegaly of two fingers' breadth under rib line spleen not palpable Other examinations were normal

The liver went down to normal size two days after admission and the icterus disappeared clinic[illegible]
was discharged on May
insulin daily Colecyst[illegible]

propamide treatment was normal, and the patient has remained in good health up to his last examination (August 23, 1958) flocculation tests, negative, bilirubin determinations normal, and Bromsulphalein retention at 45 minutes 3 6 per cent (normal, less than 6 per cent) We were not able totally to exclude the possibility of a hepatitis of virus origin, frequent in our environment but from the time of appearance, the cholangiolitic character the brief duration and the subsequent course we are inclined to interpret the case as an icterus of toxic origin (TABLE 15)

Laboratory Determinations

Hanger reaction The hanger reaction was determined in 23 patients In 21 it was negative and remained so throughout tests made from 20 to 210 days after beginning treatment In 2 cases it was positive and persisted from the beginning for 27 and 28 days respectively Suspension of treatment was considered for these two patients

Alkaline phosphatase This enzyme was studied in 21 patients In 18, it remained normal between 30 and 210 days of treatment in 2 cases it increased moderately, but became normal at 20 and 28 days, respectively, and it increased considerably in 1 case (cholangiolitic icterus)

Bromsulphalein retention Thirty seven patients were studied in this connection In 27 cases the results were normal (less than 6 per cent) between days 30 and 210 of treatment In 3 cases in which the percentages were moderately increased before initiation of treatment, values became normal within 52 and 70 days Three cases in which Bromsulphalein retention had been increased did not change during a course of treatment lasting 56 to 120 days In 3 cases that were normal before therapy, very moderate increases occurred in 65 to 107 days (8 3 to 7 8 to 10 4 per cent)

Prothrombin This factor was studied in 27 cases In 25 cases, it remained normal between 30 and 210 days In 1 case there was a moderate decrease at 84 days (without alterations in other tests) In 1 case there was a decrease that lasted for 7 days before treatment was suspended because of an intervening illness

Hemogram The blood picture was studied in 47 cases In 3 cases we observed moderate but insignificant decreases in the erythrocytes between 30 and 105 days of treatment In 1 case there was a very moderate decrease in leukocytes (4200) after 60 days of treatment

Urine Alterations were observed in only one case, where albuminuria (0 64 gm per cent) appeared after 80 days The interpretation of this case is difficult In the 5 cases in which albuminuria had existed previously, there was no modification up to 30 to 120 days of treatment

Discussion

The most significant facts encountered in this clinical test of chlorpropamide are the evidence of greater activity of this new sulfonylurea and the increased frequency of secondary reactions when the minimal effective dose is exceeded Other aspects of clinical response are the same as those observed with the use of similar substances, as may be deduced from analysis of data

in relation to age, nutritional status, and the other variable considerations always taken into account in the oral treatment of diabetes mellitus

The greater activity of chlorpropamide is principally a consequence of the fact that it rapidly reaches effective levels in the blood, probably a result of its being excreted slowly This also explains the general symptoms observed when doses higher than necessary are used, as found with the first patients we treated The digestive symptoms, on the other hand, especially the epigastric distress, can be attributed to a definite direct irritating action that probably comes from the presence of chlorine in the molecule It should be noted that most of these manifestations are only of mild or moderate intensity and of short duration, they usually disappear even with continuation of therapy From a practical viewpoint, these manifestations do not constitute obstacles to the clinical use of the drug except for an occasional patient who in our experience, usually is also a therapeutic failure

Examinations of blood and urine and tests of hepatic function revealed no significant modifications in observation periods lasting up to 210 days The only serious complication might be represented by a case of hepatitis that rapidly ran its course since that time no sequelae have become obvious

Like the other sulfonylureas, chlorpropamide is active within a few hours [illegible]

alcohol is ingested during therapy Apart from the practical interest and importance such a fact has in areas where the consumption of a moderate and permissible amount of alcohol is habitual with meals, there is a theoretical consideration in that it reveals another aspect of the action of the sulfonyl ureas, one by which they directly modify the intermediate metabolism, acting on the enzymatic processes

[illegible]

serve only to vary the absolute figures for successful results to a minor degree, at the same time still permitting comparisons to be made among various drugs

Summary

Chlorpropamide was administered to 80 diabetics (38 males and 42 females) Two of these patients were less than 20 years old, and 64 were older than 40 years 48 were obese 14 of normal weight, and 18 underweight. The diet contained 150 to 250 gm, maintenance dosage ranged from 125 mg to 1 0 gm Patients were observed from 30 to 210 days With the initial dose, the immediate hypoglycemic effect was studied on a fasting state The greatest decrease in the blood sugar level took place at 5 hours in cases that responded (selection test)

Disappearance of glycosuria and lowered blood sugar levels were obtained in 58 per cent of the cases in the first week and in 31 per cent in the second

week Results of treatment were good in 55 per cent, fair in 29 per cent, and poor in 16 per cent In the group exhibiting good results, 75 per cent were observed for more than 60 days, 66 per cent of the cases were maintained on a dosage of 500 mg or less (plasma chlorpropamide ranged between 4 6 and 9 8 mg /100 ml)

Results were correlated directly with nutritional status, age of the patient age at onset of the diabetes, and absence of previous treatment An inverse correlation was found to duration of treatment and dosage of insulin No correlation was found to duration of diabetes mellitus

There were secondary reactions in 24 cases including skin (3), digestive (20), and general manifestations (15) Such symptoms were minor in 72 per *cent and disappeared spontaneously after a decrease in dosage or after a short* period of discontinuation (6 cases) With a dosage of 1 gm , 21 per cent of reactions were observed as compared with 38 4 per cent when 2 gm was given One of the patients experienced jaundice of cholangiolitic type after 18 days of therapy Treatment was discontinued Alcohol ingestion induced anxiety, palpitations and flushes in 10 cases No changes were found in liver, kidney, or blood forming functions during or after treatment

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CLINICAL STUDY OF THE EFFECTS OF CHLORPROPAMIDE ON NORMAL SUBJECTS AND ON DIABETICS

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Since the year 1955 when Franke and Fuchs published a paper in which they recommended the use of the sulfonylurea carbutamide (1 sulfanilyl 3 butylurea or BZ 55) for the oral treatment of diabetes mellitus there have been numerous articles describing clinical experiments conducted with other compounds including *p* aminobenzenesulfamidyl *tert* butyl (RP 2259) and tolbutamide (1 butyl 3 *p*-tolylsulfonylurea or D 860) At present tolbutamide appears to be preferred by many researchers

In the hope of finding a product that might be useful to all diabetics and have effective hypoglycemic action and a very low index of toxicity research has continued in that direction lately there have been tested some derivatives of guanidine especially phenethylformamidinyliminourea (DBI) and very recently, a new sulfonylurea 1 propyl 3 (*p* chlorobenzenesulfonyl) urea (P 607) commonly referred to as chlorpropamide

The purpose of this article is to review the results achieved through the use of chlorpropamide (P 607) in persons who showed no disorders of carbohydrate metabolism and in those with diabetes It was administered to both groups for evaluation of its hypoglycemic activity and for determination of its possible untoward or toxic effects and its therapeutic usefulness

Plan of Study

Material and Methods

Thirty hospitalized patients who evidenced no disorders of carbohydrate metabolism and 60 patients with diabetes mellitus from our private practice

TABLE 1
PRIVATE PATIENTS

Total patients studied 60 outpatients 56 inpatients 4
Sex male 29 female 31
Age average 51 (range 6 to 76)

Initially with other hypoglycemics 20
Initially with insulin 34
Patients in more than one group 32
Subjects of hormone studies 27

the data on whom are compiled in TABLE 1, were selected for this research which covered a period of 90 days In the course of this work the following studies were made

Studies in Nondiabetic Subjects Without Disorders of Carbohydrate Metabolism

With a view to determining the absorption and elimination curve of the compound and the ratio between the blood concentration reached and the hypoglycemic effect obtained the subjects of this study were 30 inpatients (10 males and 20 females) who evidenced no metabolic disorder with respect to carbohydrates and who were divided into 3 similar groups of 10 patients each

After prior stabilization on a prefixed diet (total caloric intake 1833 5 cal carbohydrates 35 per cent 650 cal 162 5 gm , proteins 21 per cent 396 cal 99 gm fats 44 per cent, 787 5 cal 87 5 gm) the subjects were respectively administered a single dose of 0 5 or 1 5 gm chlorpropamide In all of the patients the blood glucose concentration was determined and in 20 of them that of chlorpropamide in the fasting state and 4 hours later in the manner described by Camerini Dávalos *et al* (1957) The same determinations were repeated in 2 subjects from each group after 8 12 48 and 96 hours

Studies of Diabetics

Studies were made of 60 patients classified in 9 different groups with a view to investigating various aspects of the problem most of them were studied from more than one aspect and therefore appear in more than one group This accounts for the apparent disparity between the actual number of patients studied and the number resulting from totaling the members of the different groups

During the research a quantitative diet was indicated for all of the patients the general characteristics of which were as follows 150 to 200 gm of carbohydrate and 75 to 105 gm of protein supplemented by fats in sufficient quantities to attain the total caloric intake desired for each particular patient

Described below is the series of 5 biochemical determinations made in addition to the findings of complete clinical examinations conducted with a view to ascertaining the evolution of diabetes and verifying at an early stage any disorder that might occur in the hematopoietic system or in hepatic and renal function (1) glycemia by Folin and Wu s serum method modified for the determination of true glucose (2) uremia by Sagastume and Oliva s gasometric method (3) complete examination of urine, (4) peripheral hemogram and (5) hepatogram including Hanger s test thymol turbidity Kunkel s test alkaline phosphatase cholesterolemia bilirubinemia fractional proteinemia and electrophoresis on paper

Group I This group was composed of 10 patients who initially received a dosage of chlorpropamide for 48 hours of 1 5 gm /day divided into 2 or more doses followed by 1 gm day for not less than 15 days For the purpose of verifying the occurrence of untoward symptoms and gastrointestinal tolerance these patients also received at various times and under different circumstances antacids proportioning of doses and administration concurrently with ingestion of food

In 5 patients the drug was beneficial and dosage was reduced to a minimum

effective level These patients were added to the group of diabetics receiving protracted treatment

Group 2 This group included 20 diabetics who were well balanced with other hypoglycemic compounds 8 received carbutamide and 12 received tolbutamide (TABLE 2) The initial dose of chlorpropamide which was started *immediately after suspension of previous therapy* was *1 gm /day* for 2 days and we attempted to reduce it rapidly to lower levels according to the therapeutic response obtained The purpose of this procedure was to make a comparative clinical study of various hypoglycemics in relation to such factors as the efficacy of a dose administered for a protracted time

TABLE 2

Age and sex (present age females are indicated in parentheses)								
Ages	1 to 10	11 to 20	21 to 30	31 to 40	41 to 50	51 to 60	61 to 70	71 to 80
	—	—	—	—	2 (1)	9 (4)	8 (5)	1 (1)
Age at onset of diabetes								
Ages	1 to 10	11 to 20	21 to 30	31 to 40	41 to 50	51 to 60	61 to 70	71 to 80
	—	—		2	8	9	1	—
Years of diabetes at start of treatment								
Years	Recent to 1		1 to 2	3 to 5	6 to 10	11 to 15	16 to 20	21 to 36
	—		4	5	1	6	3	1
Body type pyknic *1* normal *18* longilineal *1*								
Type of diabetes stable 20 (100%) unstable 0								
Prior drug therapy		0 5			1 gm			1 5 gm
D 860		—			9			3
BZ 55		4			4			—
Duration of prior treatment								
	Less than 6 months			6 months to 1 year			1 to 2 years	
	4			8			8	
Prior control good 19 fair 1								

tolerance and the continuance of proper regulation after suspension of the drug

In all of these patients chlorpropamide therapy was continued for a prolonged period Five patients from group 1 and 16 from group 4 who also continued to receive the drug joined them thereby raising to 41 the total number of subjects in whom the effects of the compound administered for a

Group 3 This group comprised 9 patients in whom carbutamide and tolbutamide had been inefficacious These patients had received insulin and were selected with a view to drawing clinical conclusions on the similarities between the different compounds The group includes one case that had presented a good *response but that was complicated by skin eruption* and icterus

Group 4 In this group were 34 cases who had never before been treated with oral hypoglycemics, in 8 of whom no improvement had been produced by dietary restriction, and who had required insulin for years for the proper regulation of their diabetes The data on the members of this group are set forth in TABLE 3

When insulin requirements had been lower than 30 U, insulin therapy was suspended, and treatment with an average dose of 1 gm of chlorpropamide was started immediately On the other hand, in those patients who had received more than 30 U, along with the beginning of treatment with hypoglycemics (125 to 250 mg /day), the dose of which was then gradually augmented, the dosage of insulin was scaled down by a percentage equivalent to 20 per cent of the previous dose every 3 or 4 days

TABLE 3

Age and sex (present age females are indicated in parentheses)

Ages	1 to 10	11 to 20	21 to 30	31 to 40	41 to 50	51 to 60	61 to 70	71 to 80
	3 (3)	3 (1)	1	2 (1)	5 (1)	10 (6)	8 (6)	2 (2)

Age at onset of diabetes

Ages	1 to 10	11 to 20	21 to 30	31 to 40	41 to 50	51 to 60	61 to 70	71 to 80
	5	1	3	4	8	10	2	1

Years of diabetes at start of treatment

Years	Recent to 1	1 to 2	3 to 5	6 to 10	11 to 15	16 to 20	21 to 36
	1	5	12	10	2	3	1

Body type pyknic 2, normal 28, longilineal 4

Type of diabetes stable 25 (75 per cent), unstable 9 (25 per cent)

Prior dose	30 U or less	More than 30 U
	18	16

Duration of prior treatment

Less than 1 year	1 to 5	6 to 10	11 to 20	More than 20 years
—	7	11	12	4

Prior control good 24 fair 10 poor —

Group 5 In this group were 6 patients who were well regulated by dietary restriction alone When under therapy, these patients were permitted a free and ample diet or prescribed 300 gm of carbohydrates daily for the purpose of observing to what extent a sustained dose of chlorpropamide ranging from 125 to 500 mg would make possible an ad libitum diet. They were administered the compound for periods of 30 days or more

Group 6 This included 2 infant diabetics and 3 juvenile unstable diabetics, in whom chlorpropamide was tested in doses ranging from 125 mg to 1 5 gm, at the same time we attempted to reduce the insul[illegible] minimum

Group 7 Th[illegible] function by t[illegible] compounds had already been s[illegible] of the determination of serum protein bound iodine (Barker and Humphrey's method), of basal metabolism, and of I[131] uptake observed on the gland after 2 and 24 hours, the possible influence that might be exerted by chlorpropamide, when administered over a protracted period in an average dosage of 1 gm daily, was studied

Group 8 The following determinations were made in 10 diabetics, with a view to checking any disorder that might occur in hypophyseal and supra-

renal endocrine function hypophyseal gonadotropins by Albright Reifenstein and Griswold's method urinary neutral 17 ketosteroid by Callow, Gallow and Emmen s method and urinary 17 hydroxycorticoids by Reddy Renkins and Thorn s method

Group 9 This was a heterogeneous group of 8 diabetic patients who manifested the special conditions or associated endocrinopathies listed below

These patients were administered chlorpropamide in varying dosages for the purpose of observing the changes that might occur in the existing equilibrium with respect to the carbohydrate metabolism insulin resistance 1

In the last 4 groups control determinations were made whenever it was possible to establish a minimum effective dosage gradually reached after initial doses of 250 to 500 mg

Results and Their Interpretation

Studies of Nondiabetic Subjects

The results obtained in this study are tabulated in TABLES 4 and 5 in which have been compiled all of the blood glucose concentrations and chlorpropamide values expressed in milligrams per 100 ml of blood

From study of the values transcribed it can be concluded that chlorpropamide when administered orally in humans is rapidly absorbed and reaches efficacious blood concentrations in a few hours The level attained as far as the doses used by us are concerned remained in direct ratio to the quantity administered and was sustained with proportionally the same differences during the first 48 hours After 96 hours there was a marked decline in the values which in 4 of 5 cases observed failed to show any significant differences from one another The decline registered in the glycemic values was quite pronounced within 4 hours after ingestion since the chlorpropamide concentration had by that time reached at least 2 5 to 3 5 mg 100 ml we believe these concentrations are the minimum levels for therapeutic efficacy The hypoglycemic action remained evident for the first 24 hours diminishing during the following hours despite the fact that there were effective blood levels a fact we ascribe to the alimentary ingestion after the first day of observation

It must be emphasized that in this experiment the glycemic decline was always pronounced but did not remain in direct proportion to the blood concentration attained by the drug since the latter had exceeded effective blood levels (greater than 3 5 mg /100 ml)

Studies of Diabetics

Group 1 Seven patients of 10 received high doses for several days and 12 took a high dose only initially Aside from the hypoglycemic effects produced in these patients untoward side effects were manifested (TABLE 6) and on various occasions some of them experienced more than one reaction

TABLE 4

Dose of chlor prop-amide	Case No	Glucose mg /100 ml		P 607* mg /100 ml 4 hr	Dose of chlor prop-amide	Case No	Glucose mg 100 ml		P 607 mg 100 ml 4 hr	Dose of chlor prop-amide	Case No	Glucose mg 100 ml		P 607 mg /100 ml 4 hr
		Fasting	4 hr				Fasting	4 hr				Fasting	4 hr	
	1	94	73	—		11	92	68	—		21	100	70	10 8
	2	78	56	2 8		12	83	62	7 7		22	89	80	—
	3	100	84	1 5		13	83	64	8 4		23	87	77	10 4
	4	92	61	—		14	121	78	—		24	94	72	—
	5	118	69	2 5		15	75	67	—		25	90	60	11
0 5 Gm	6	90	70	4 6	1 Gm	16	113	70	10 2	1 5 Gm	26	120	90	9 4
	7	90	75	—		17	97	—	8 8		27	91	68	10 4
	8	97	68	1 5		18	76	74	8 2		28	77	57	9 6
	9	91	77	—		19	96	72	8 6		29	95	62	10 6
	10	82	69	—		20	103	75	6 7		30	74	55	9 2

* P 607 = chlorpropamide

TABLE 5

Dose of chlorpropamide	Case No	mg /100 ml	Fasting	4 hr	8 hr	12 hr	24 hr	48 hr	96 hr
0 5 Gm	5	Glucose	118	69	60	70	65	93	92
		Chlorpropamide	—	2 5	5 6	3 3	—	4 8	2 3
	6	Glucose	90	70	93	81	65	90	96
		Chlorpropamide	—	4 6	2 1	3 1	—	2 3	1 5
1 Gm	17	Glucose	97	—	103	72	89	94	108
		Chlorpropamide	—	8 8	5 2	5 7	—	4 8	1 9
	18	Glucose	76	74	75	77	64	101	112
		Chlorpropamide	—	8 2	5 9	5 9	—	5 4	3
1 5 Gm	21	Glucose	100	70	98	88	83	103	100
		Chlorpropamide	—	10 8	12 1	9 0	—	8 8	3 1
	23	Glucose	87	77	47	63	68	90	—
		Chlorpropamide	—	10 4	10 1	10 1	—	7 3	—

TABLE 6

TOTAL UNTOWARD SIDE EFFECTS OBSERVED

Facial flushing and feverishness 9
Skin reactions 4
Digestive disturbances (pyrosis regurgitation bloated feeling anorexia) 10
Constipation occasional impossible to determine accurately
Headache 3
Feeling of general malaise with vertigo 2
Increase in blood pressure? 2

It is to be pointed out that reactions of the vasomotor type accompanied by feverishness and facial flushing frequently broke out after ingestion of an alcoholic beverage, spontaneously receding within 30 min and failing to occur when the dose of the drug was reduced or the ingestion of alcohol was suspended

The skin reactions were largely minor eruptions and on two occasions, became evident after the administration of a saline laxative In two patients it was necessary to discontinue the drug, application of which could be resumed without side reaction only in smaller doses once the cutaneous episode had subsided

Gastrointestinal disorders occurring largely in the upper digestive tract were the disturbances most persistently complained of by the patients

These manifestations could be relieved with the administration of antacids and proportioning of dosage of the drug prescribed and by the ingestion of both at meal times, but *the disorders could not be totally eliminated* until the dosage was reduced below 1 gm /day

Some patients complained of constipation, occasional headache, and malaise and showed an increase in blood pressure, all of these disturbances were transitory, and their cause and *effect relationship with chlorpropamide cannot be defined by the present study* Investigations of liver function and blood count were unsuccessful in relating any of the above mentioned disturbances to a possible attack on the hepatic parenchyma or hematopoietic system Analyses of blood and urine urea did not show any significant changes

Five patients in this group subsequently continued therapy with doses of chlorpropamide not exceeding 750 mg /day

Group 2 In none of the 20 patients who had been kept well regulated with the use of carbutamide or tolbutamide was any disturbance observed when the previous therapy was suspended and chlorpropamide immediately substituted *Chlorpropamide proved efficacious in all of these cases*

tolbutamide the effective dosage of chlorpropamide was determined to be *3 times (in some instances 4 times) smaller than that of the hypoglycemic* previously used

Group 3 In 4 of 9 patients in whom other oral hypoglycemics had been ineffective the same poor results were observed when chlorpropamide was used in the other 5 cases *chlorpropamide produced satisfactory regulation in contrast to the results previously obtained* with carbutamide and tolbutamide It should be pointed out that in 2 of the latter patients the failure of the drug previously used was due partly to untoward side effects that contraindicated their use (severe icterus) It should also be noted that on 2 occasions patients well balanced with chlorpropamide required insulin *during the acute period of an intercurrent febrile process*

Group 4 In 8 cases receiving an insulin dose of less than *30 U*, it was possible to suspend insulin completely and replace it with chlorpropamide On the other hand only 5 patients receiving more than *30 U* of insulin daily could forego the injections

In 6 cases the insulin dosage, which had ranged between 40 and *70 U*, *could be significantly* reduced (by 20 per cent or more) when chlorpropamide

up this group *the addition of chlor* it possible to broaden the diet to *normal limits* without untoward effects It must be pointed out, however, that repeated dietetic transgressions or excessive gain in weight contra indicated the continuation of this leniency in the majority of the patients

Group 6 In none of the 5 members *of the group was it possible to suspend* insulin entirely however, in 2 cases it was possible to reduce *the dose by*

50 per cent of the previous quantity when chlorpropamide was also administered in quantities of 1 and 1 5 gm /day respectively In a third case the reduction of the insulin dose was equivalent to only 10 per cent of the previous dose but perfect metabolic equilibrium was obtained without hypoglycemia which until then had recurred daily

Group 7 In none of the 17 patients making up this group were there any clinical manifestations attributable to hypothyroidism The variations in values obtained after administration of the drug for long periods and in doses

TABLE 7
THYROID STUDY

	I^{131} (percentages)		Serum protein bound iodine (µg)	Basal metabolism (per cent)	Observations
	2 hr	24 hr			
1	27	47	3 3	+ 1	
2	11	4	1 9	+14	Hypothyroidism (under treatment)
3	25	47	3 3	+33	
4	20	44	4 1	− 3	
5	23	44	3 7	± 0	
6	17	36	2 6	+18	
7	20	53	3 6	+ 8	
8	19	31	4 1	−10	
9				+25	
10				− 2	
11	41	62	9 1	+30	Hyperthyroidism
12	33	44		+ 0	
13	34	56	8 0		Hyperthyroidism
14	10	34	3 1	+40	
15				−12	
16				− 9	
17	15	32	3 7	+ 5	

[illegible] ference in the sense of func [illegible]erted in 8 o[illegible] reaching statistically significa[illegible] a slight tendency to diminish in value (TABLE 7)

Group 8 The results of the hormonal determinations made in this group of patients and summed up in TABLE 8 do not make it possible to state that chlorpropamide exerts any influence on the glandular functions studied

Group 9 Among the 8 patients with special conditions, in the patient who demonstrated resistance to insulin it was possible to reduce the insulin dose from 500 to 60 U /day when chlorpropamide (2 gm /day) was added to the treatment, this drug also contributed to greater metabolic stability

and relieved the patient of the recurrent periods of chemical acidosis from which he constantly suffered

The 3 patients with allergy to insulin tolerated this new substance without incident In the patient with the adrenogenital syndrome, in spite of the extraordinarily high content of urinary 17-ketosteroids (120 mg in urine after 24 hours), the response to chlorpropamide was similar to that registered in the cases without endocrinological complications

TABLE 8
URINARY DETERMINATION OF HYPOPHYSEAL GONADOTROPINS, NEUTRAL 17 KETOSTEROIDS AND 17 HYDROXYOXYSTEROIDS

Case No	Gonadotropins		17 K S mg / 24 hours	17 H K S mg / 24 hours	Observation
	To 6 U L	To 96 U L			
1	(+)	(−)	7 20	2 63	—
2	(+)	(−)	—	—	—
3	(+)*	(−)	6 44	1 68	—
4	(+)	(−)	—	—	—
5	(+)*	(−)	4 68	4 45	—
6	(+)	(−)	5 40	6 98	—
7	(+)	(+)*	1 20	2 19	Adrenogenital syndrome
8	(+)	(+)	4 62	7 98	Menopause
9	(+)	(+)	8 30	3 41	Menopause
10	(+)	(−)	6 21	4 82	—

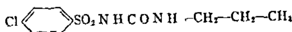
* Weak

In both patients with functional disturbance of the thyroid gland it was extremely difficult to obtain adequate control, as had been the case with all of the treatments previously employed, because of the accentuated metabolic instability with marked ketosis trend constantly present

In the leprous patient the response obtained with chlorpropamide was to be expected, taking into account solely his metabolic disorder

COMMENTS

Chlorpropamide is a compound very similar in chemical structure to carbutamide and tolbutamide,



having in common with them the part included in the box drawn with broken lines, but differing in that it has a propyl radical instead of butyl, and that in ' – -- pos t on of the benzene nucleus it has a chlorine instead of the an

the absence of the amino radical in chlorpropamide means that it has no bacteriostatic action for which reason undesirable changes in intestinal flora are not registered when it is used This fact has been corroborated by this study On the other hand the aforementioned disorders of the upper gastrointestinal tract become common (in about 70 per cent of the subjects studied) when daily doses of more than 1 gm are administered In no case did we witness such effects when the dosage did not exceed 750 mg / day

The curves pertaining to nondiabetics reveal the rapid absorption of the compound and its marked therapeutic efficacy in doses of 0 5 gm and even less since after 4 hours the glycemic levels had descended by at least 20 per cent with respect to the initial levels The greater quantities resulted in a higher blood concentration of the drug and in its slower elimination from the blood stream (good concentrations after 96 hours) which accounts for the protracted therapeutic effect after suspension of the drug and for the high concentrations attained with repeated doses These observations coincide with other experimental and clinical findings (Goldner 1958) to the effect that effective blood levels attained after 48 hours slowly drop on suspension of the drug until its complete elimination from the blood stream within a period of 5 to 7 days According to Heinsen (1958) 50 per cent of an oral dose will still remain in the blood after 24 hours This opinion is shared by Miller (1958) according to whose experience with a dose of 1 gm /day the blood concentration rises gradually for a period of 1 week and is then maintained at the last levels reached

In one of our cases in which the maintenance dose was 1 5 gm day 25 days after treatment the blood chlorpropamide concentration was 16 5 mg /100 ml in 3 other patients receiving 750 mg over the same period the values were 8 4 10 8 and 13 3 mg /100 ml respectively In one of the patients who received 500 mg /day the figure was 9 4 mg ml on the other hand in 2 other cases with the same dose the values were 4 6 mg and 5 4 mg /100 ml which reveals the importance of the size of the dose by itself and the variations that can arise according to the individual susceptibility of each patient

Taking the foregoing into account and also the fact that the hypoglycemic effect bore no direct relationship to the dosage since effective therapeutic levels had been attained it does not seem hazardous to affirm that the quantities indicated for clinical use should not exceed 750 mg /day whether in single or proportioned doses the more so since the risks of hypoglycemia and the onset of untoward side effects become more common with higher doses

Without being able to point to a definite pathogenic mechanism (perhaps the inhibition of alcoholic dehydrogenation) mention should be made of the frequency with which we observed a reduced alcoholic tolerance manifested by feverishness and facial flushing, which ceased with the reduction of the dose or suspension of alcohol

The possibility of satisfactorily regulating diabetics who had previously been receiving carbutamide or tolbutamide successfully was confirmed in 100 per cent of the cases studied in such connection and it must be added that chlorpropamide was also efficacious in cases that had not responded to

other drugs This therapeutic efficacy becomes evident with doses that are usually 3 or 4 times smaller than those of carbutamide and tolbutamide

In almost all of our case histories the effective quantity of chlorpropamide for prolonged treatment was less than 1 gm /day, in many instances only 0 5 gm , and in several cases 0 25 gm and 0 125 gm /day were efficacious This fact bears out the observations made by Root (1958) in the rat, when she showed that chlorpropamide was 20 per cent more active in this animal than carbutamide and 60 per cent more efficacious than tolbutamide

In all of our cases the blood chlorpropamide concentration was maintained at effective levels and, when the doses were 1 gm or larger, the figures rose significantly, as already mentioned with reference to the results In no instance was any definite attack on the hepatic or renal parenchyma or hematopoietic system evidenced although, on two occasions, positive liver flocculation tests and the elevation of alkaline phosphatase observed in patients maintained for a relatively long time on 1 gm of the compound raised a question as to the harmlessness of the substance to the parenchyma of the liver when administered over a protracted period of time in high dosage (see results among 15 patients in TABLE 9)

In general the good therapeutic responses in our patients maintained the same relationship with various factors as those reported by Menhert *et al* (1958) with respect to tolbutamide the onset of disease in middle age, small previous insulin requirements, short history of diabetes, and recent insulin treatment

The foregoing bears out the therapeutic inefficacy observed in infantile and juvenile patients and in several of those patients who required insulin doses of more than 30 U , in all of whom the continuation of treatment with both drugs seems indicated only when necessary to attain metabolic stability that cannot be achieved by other means, since not only have we been able to observe a diminution of broad glycemic variations, but also a marked "warding off" of ketosis, which in several cases was readily manifested when insulin alone was used

In our cases the reduction of insulin dosage among patients who had depended exclusively on this hormone for regulation ranged between 10 and 100 per cent This bears out similar observations made in humans with respect to tolbutamide by Mirsky and Diengott (1957) and by Fabrykant (1958), which confirm those carried out in the rat with tolbutamide by Houssay *et al* (1957) and with chlorpropamide by Root (1958)

The increased metabolic stability offered by some oral hypoglycemics has also been pointed out with special reference to carbutamide by Izzo (1957), to tolbutamide by Fulner *et al* (1956), Bander (1956), Fabrykant (1958), and to various biguanidines by Krall and White (1958)

It is interesting to note that several patients who were controlled previously with both insulin therapy and later with chlorpropamide claimed that they felt better with the latter substance This fact was more pronounced in children, even in periods of unsatisfactory compensation

This feeling of good health made it necessary to be extremely vigilant as to observance of the prescribed diet, for patients might easily commit serious

TABLE 9

Case No	Hanger test	Thymol turbidity	Kunkel test	Alkaline phosphatase	Cholesterol	Bilirubin	Proteins	Electrophoresis	Hemogram
1	(–)	2 1	5 2	3	262	D = 0 05 I = 0 55 Tot = 0 6	A = 4 46 G = 2 23 Tot = 6 69 A G = 2	A = 3 01 = 45% α_1 = 0 27 = 4 1% α_2 = 0 80 = 12% β_1 = 0 80 = 12 1% β_2 = 0 35 = 5 3% γ = 1 43 = 21 5%	
2	(–)	0 5	2 5	2 64	260	D = 0 1 I = 0 5 Tot = 0 6	A = 4 62 G = 2 61 Tot = 7 23 A G = 1 76	A = 3 75 = 51 9% α_1 = 0 45 = 6 3% α_2 = 0 74 = 10 3% β = 0 93 = 13% γ = 1 31 = 18 2%	
3	(–)	4	6	16 4	202	D = 0 I = 0 4 Tot = 0 4	A = 4 G = 2 46 Tot = 6 46 A G = 1 62	A = 3 30 = 51 2% α_1 = 0 27 = 4 3% α_2 = 0 73 = 11 4% β = 0 74 = 11 5% γ = 1 38 = 21 4%	
4	(–)	3 5	6	2 68	250	D = 0 1 I = 0 6 Tot = 0 7	A = 4 46 G = 2 54 Tot = 7 A G = 1 71	A = 3 74 = 53 4% α_1 = 0 29 = 4 18% α_2 = 0 71 = 10 12% β = 0 66 = 9 43% γ = 1 60 = 22 85%	

TABLE 9 (*Continued*)

Case No	Hanger test	Thymol turbidity	Kunkel test	Alkaline phosphatase	Cholesterol	Bilirubin	Proteins	Electrophoresis	Hemogram
5	(−)	2	4.5	6.18	242	D = 0.15 I = 0.65 Tot = 0.80	A = 4.31 G = 2.77 Tot 7.08 A/G – 1.55	A 3.49 = 49.3% α = 0.22 = 3.2% α_2 = 0.62 = 8.8% β = 1.17 = 16.6% γ = 1.55 = 21.9%	
6	(−)	3.1	5.8	10.1	216	D = 0 I = 0.5 Tot – 0.5	A – 4.64 G = 2.90 Tot – 7.54 A/G = 1.60	A = 3.55 = 47.2% α = 0.23 = 3.1% α_2 = 1.17 = 15.6% β = 0.98 = 13% γ = 1.57 = 20.9%	
7	(−)	4.5	10	3.5	230	D = 0.15 I = 0.70 Tot = 0.85	Tot = 6.55		R B C 4 700 000 W B C 5 900 Hb 98%
8	(−)	5	6	1.46	290	D = 0.1 I = 0.58 Tot = 0.68			R B C 5 100 000 W B C 6 900 Hb 92% Diff N = 59% I = 7 B = 0 L = 30 M = 4
9	(+)	4.2	5	2.33	260	D = 0.05 I = 0.61 Tot = 0.66			R B C 4 640 000 W B C 5 900 Hb 76% Diff N = 62% I = 4 B = 0 I = 30 M = 2

10	(++)	2 05	4 5	5 38	230	D = 0 05 I = 0 50 Tot = 0 55			R B C 5 080 000 W B C 6 800 Hb 96%, V G = 0 94 Diff N = 72% I = 3 B = 0 I = 22 M = 3
11	(++)	3 8	4 2	2 92	315	D = 0 1 I = 0 68 Tot = 0 78			R B C 5,050 000 W B C 10 000 Hb 14 5 G Diff N = 61% I = 1 B = 0 L = 33, M = 5
12	(+++)	17 7	2 3	8 49	1 95	D = 0 15 I = 0 75 Tot = 0 9			
13	(+++)	5 6	8 5	5 21	206	D = 0 1 I = 0 59 Tot = 0 69			R B C 5 580 000 W B C 8 100 Hb 16 2 G = 108% V G 0 93 Diff N = 62% L = 0 B = 0 L = 31 M = 5
14	(−)	1 5	2	3	212	D = 0 I = 0 6 Tot = 0 6			R B C 4 900 000 W B C 7 000 Hb 12 8 G = 86% Diff N = 69% E = 2, B = 0 L = 26, M = 3
15	(+)	3 5	7 2	1 39	262	D = 0 05 I = 0 64 Tot = 0 69			R B C 4 700 000 W B C 6 400 Hb 12 6 G = 84% Diff N = 67% I = 1 B = 0 L = 28 M = 4

BLOOD LEVELS OF CHLORPROPAMIDE IN NORMAL MEN FOLLOWING CHRONIC ADMINISTRATION

Michael Carlozzi, Domenic G Iezzoni, Lawrence Silver
Chas Pfizer & Co, Inc, Brooklyn, N Y

As part of a large study program on the efficacy and activity of the new oral hypoglycemic agent chlorpropamide, it was felt that a knowledge of the blood levels at various dosages following chronic oral administration would be of importance We were already aware that only small amounts are excreted in the urine Therefore it seemed necessary to determine whether serum levels continued to rise with repeated administration or whether some plateau value would ensue

Method

Three groups of normal male volunteers were placed on a daily dosage schedule of either 0 25, 0 50 or 1 0 gm chlorpropamide daily The group

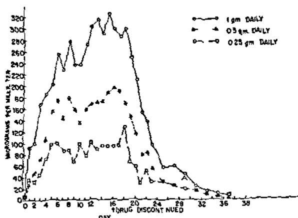


FIGURE 1 Chlorpropamide serum levels

taking the highest dose was composed of 20 men, the other 2 groups comprised 10 men each A fasting blood sample was drawn on the morning of the first day and the drug was administered orally immediately afterwards

received 100 mg daily The entire group on 1 0 gm daily had the drug discontinued at the end of 15 days, and the serum levels were followed through the thirty seventh day Usual diets were maintained throughout the study period except for added carbohydrates as needed to relieve sensations of weakness and hunger Analyses of the serum for chlorpropamide were carried out in our laboratories by the method of Toolan and Wagner[1]

TABLE 1
BLOOD LEVEL OF CHLORPROPAMIDE 0 25 GM DAILY (μg /ml)

Patient	Day 1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	25	27	29	31	33	35	37
21	63	30	41	43	95	87	35	43	35	77	43	90	77	40	63	80	75	75	48	47	18	37	10	0	35	42	0	0	0	0
22	44	30	40	83	–	75	90	375	—	—	—	—	—	50	48	75	82	250	55	63	25	48	32	—	—	15	30	—	10	10
23*	55	41	47	78	125	135	120	130	85	112	103	185	158	110	160	138	207	130	125	122	90	110	80	15	77	50	40	32	25	20
24	45	49	45	48	98	100	77	80	60	75	58	100	75		85	93	113	—	—	—	—	—	—	—	—	—	—	—	—	—
25*	43	42	76	74	113	95	88	130	100	162	92	125	90	123	100	120	110	123	110	125	90	125	50	40	50	50	35	32	25	20
26*	46	52	52	67	110	115	127	118	70	145	98	107	93	113	105	123	123	105	102	85	70	82	68	—	50	55	45	55	—	25
27	39	43	41	73	92	110	83	92	70	85	65	50	80	90	97	98	83	82	60	47	30	50	30	85	15	10	18	20	—	10
28	39	44	68	97	117	103	90	120	40	85	90	100	90	140	123	123	125	107	102	72	38	67	48	10	18	10	—	12	0	0
29*	40	44	69	62	50	87	75	77	60	80	75	75	60	87	102	113	128	83	55	70	58	70	57	48	42	15	30	20	10	10
30*	43	40	64	73	85	92	75	100	77	77	75	70	65	98	68	102	85	70	70	48	18	62	47	—	15	40	0	0	0	0
														Mean no drug		94	96	129	66	57	28	51	30	32	23	19	16	11	3	5
Mean	46	42	54	73	98	100	86	88	66	99	78	101	88	95	95	119	131	102	92	90	65	90	60	34	47	42	30	28	15	15

* Started on 100 mg daily on sixteenth day others discontinued

TABLE 2
BLOOD LEVEL OF CHLORPROPAMIDE, 0.5 GM DAILY (µg/ml)

Patient	Day 1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	25	27	29	31	33	35	37
31	15	23	48	63	93	90	85	119	98	110	95	115	110	130	150	123	115	90	90	50	30	25	0	0	0	—	0	0	0	0
32*	35	62	85	156	115	197	177	235	188	238	235	230	240	263	250	270	293	410	222	162	132	182	155	27	80	67	42	—	35	25
33*	30	63	90	122	151	190	173	220	160	185	183	200	200	195	220	250	220	180	180	152	90	125	105	62	50	35	35	—	25	20
34*	21	48	80	98	190	115	108	160	165	90	148	100	162	180	185	175	167	150	135	105	88	—	65	55	33	20	12	0	0	0
35	20	46	63	123	163	165	142	190	145	130	145	198	200	190	227	220	200	190	165	90	80	75	57	47	10	22	20	0	0	0
36	34	57	109	153	230	265	188	245	212	215	233	250	253	260	283	275	233	200	190	150	85	125	77	63	30	32	20	30	0	0
37	36	65	110	110	210	190	150	230	200	160	210	223	165	235	193	193	—	220	177	170	115	120	90	—	50	80	27	35	20	10
38*	31	49	65	84	123	125	100	127	120	112	110	105	110	—	140	145	123	112	80	90	50	75	57	42	—	50	20	50	20	15
39	30	52	78	115	226	250	157	212	240	177	170	192	205	225	187	177	220	150	115	125	90	77	57	10	30	18	—	0	0	0
40†	5	0	0	36	60	—	—	63	58	33	75	65	52	80	55	120	83	50	30	30	0	30	—	0	0	0	0	0	0	0
														Mean off drug		198	192	170	147	117	80	84	56	30	25	38	17	13	4	2
Mean	26	47	73	106	156	176	142	180	159	135	160	168	170	173	189	210	201	213	154	127	90	127	96	47	54	43	27	25	20	15

* Started on 100 mg daily on sixteenth day, others discontinued
† I xcluded from mean

Table 3

Patient	Day 1	25	27	29	31	33	35	37
1	105	30	35	30	18	—	0	0
2	85	93	62	72	32	27	15	10
3	92	40	15	35	10	0	0	0
4	85	50	45	50	30	35	10	10
5	91	40	18	25	15	0	0	0
6	100	68	70	62	35	27	20	10
7	128	—	40	60	30	20	10	10
8	88	35	30	25	40	0	10	0
9	93	50	50	50	30	20	10	10
10	79	38	40	42	15	12	0	0
11	75	90	50	48	45	40	20	12
12	82	63	47	42	18	0	0	0
13*	58	0	40	48	25	0	0	0
14*	0	12	12	30	17	10	0	0
15	77	60	65	52	15	0	0	0
16	80	65	62	35	12	0	0	0
17*	63	20	35	60	18	0	0	0
18	98	73	77	50	27	—	10	10
19	0	77	133	80	35	30	20	20
20	77	18	—	0	0	0	0	0
Mean	92	57	59	45	24	14	8	5

* Excluded from mean, a

Results and Discussion

The average daily serum levels of chlorpropamide for the three dosages administered are given in FIGURE 1. TABLES 1, 2, and 3 show that the individual serum levels within each group were spread over a considerable range

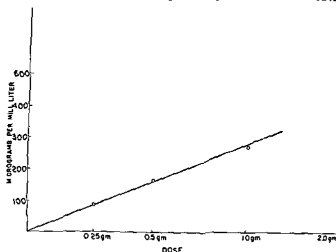


FIGURE 2. Log dose chlorpropamide serum level curve.

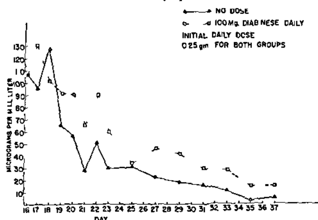


FIGURE 3. Effect of chlorpropamide—100 mg. daily—on maintenance of serum level.

of values. However, each subject, on a day-to-day basis, had reasonably consistent values. It is apparent that at 0.25 and at 0.50 gm. daily the serum levels reach a plateau in 3 to 5 days around which there is some daily variation. At 1.0 gm. daily it appears that the plateau is reached somewhat

siderably slower than that for tolbutamide and presumably, accounts for the more prolonged effect that has been reported FIGURES 3 and 4 demon

were maintained on chlorpropamide at 100 mg daily after discontinuing the prior larger daily doses as in those where the medication was discontinued

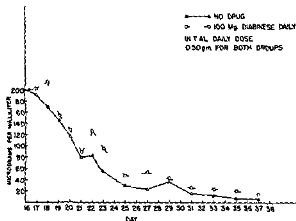


FIGURE 4

after 15 days During the course of the study there were no subjective or objective symptoms observed However many of the subjects did experience hunger and supplemented their diet with chocolate and hard candies

Summary

Various doses of chlorpropamide were given orally to normal volunteers Serum levels were followed for thirty seven days No untoward effects were observed

Reference

1 TOOLAN T & R I WAGNER JR 1959 The physical properties of chlorpropamide and its determination in human serum Ann N Y Acad Sci 74(3) 449

SULFONYLUREA DRUGS IN THE TREATMENT OF DIABETES MELLITUS

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National Institute of Nutrition, Buenos Aires, Argentina

Introduction

The National Institute of Nutrition of Argentina first began clinical investigation of the sulfonylurea drugs with carbutamide in March 1956. The results were reported at a meeting of the Argentine Association of Nutrition and Dietetics in October 1956 and were published in 1957.[1] We expanded our work to include tolbutamide, and a report of the results obtained with this drug was also published.[2] Some of us have reported separately on our experiences with the sulfonylurea drugs.[3-11] Two conferences were held on sulfonylurea drugs by the Argentine Medical Association (in 1957 and 1958) and two special sessions were held by the Argentine Association of Nutrition and Dietetics.

Theoretically, the ideal sulfonylurea drug would provide the following: (1) minimal gastrointestinal intolerances, with no acute or chronic side reactions or toxicity and an ample margin of safety, that is, a wide difference between the therapeutic dose and the toxic dose; (2) a favorable action in ... -utritional ... ercurrent ... al physiological mechanism that maintains sugar metabolism.

An analysis of all available preparations reported in the literature was published by one of us in June 1958[6] and is summarized in TABLE 1. It is evident from the ample literature of the past three years that we are far from achieving the ideal therapeutic agent. For this reason, the appearance of any hypoglycemic drug makes it mandatory that we determine how close it comes to the therapeutic ideal and establish a comparison with insulin and drugs previously evaluated in regard to efficacy, dosage, intolerance, and possible future effect on the therapy of diabetes.

With this point of view in mind the National Institute of Nutrition, the Chair of Pathology, and the Nutrition Clinic, in association with other Argentine centers for the study of diabetes situated in La Plata, Mendoza, and Córdoba carried out a study of the clinical effects of chlorpropamide. The method involved rigid conformance to the same techniques at different places, which invests this project with both a unity and a diversity that affect the significance of the results.

Table 1

Consensus on Action of and Indications for Benzenesulfonamides*

Benzenesulfonamides	Probable locus or mode of action
p aminobenzenesulfamidoisopropylthiodiazole	(1) Pancreas
	(2) Inhibition of insulin degradation
p aminobenzenesulfonylbutylurea	(3) Liver enzyme systems
p methylbenzenesulfonylbutylurea	(4) Peripheral effect on carbohydrate metabolism

Criterion of case selection	Factors indicating use of benzenesulfonamides
(1) Age	(1) More than 50 years old
(2) Duration of diabetes	(2) Less than 10 years
(3) Duration of insulin treatment	(3) Less than 5 years
(4) Insulin dosage	(4) Less than 40 U
(5) Constitutional type	(5) Pyknic, with obesity and essential hypertension tendencies
(6) Fasting blood sugar level before treatment	(6) Not more than 300 mg /cc
(7) Ketosis tendency	(7) None

* To June 1958

Material and Methods Employed

Characteristics of the Patients Studied

The drug was used on 200 patients, but data have been tabulated for only 130 these patients have been treated for a sufficient length of time to permit an evaluation to be made (figure 1)

Conditions Under Which the Study Was Conducted

Form of control Of the total of 130 patients whose data have been tabulated, 30 (23 per cent) were studied while they were hospitalized, and 100 (77 per cent) were observed in the outpatient department The latter were

selected from patients who assured us that they would follow instruction carefully. To evaluate fully the effect of the drug we administered it to patients having various types of diabetes. After observations of response were made, we continued our investigation of those individuals who had some degree of satisfactory response to the drug.

The diet followed. A nutritional study was conducted in order to acquire a knowledge of the diet of all patients. A dietary interview was held with each patient during which a detailed history of his dietary habits was recorded.

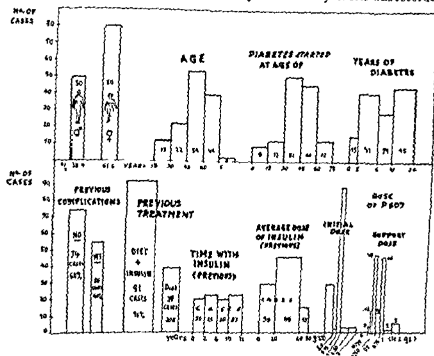


FIGURE 1. Characteristics of 130 cases under observation.

Dietary regimens, with the characteristics of these normal regimens introduced, were designed to provide each patient with the caloric value appropriate for his or her nutritional status, and to include approximately 200 gm. of carbohydrates; this amount was reduced in certain cases and increased in others to as much as 300 gm. It is understood, of course, that the other values—those of the minerals and vitamins—were the same as in the normal regimen in every case.

Examinations and tests used to evaluate the response to therapy. The observation of each patient in the study was carried out under strict clinical, medical, and laboratory control. Clinical determinations included body weight, diuresis, and any subjective or objective symptoms that appeared. Laboratory determinations included blood sugar before breakfast, blood sugar profile during the day, total daily urine sugar and urine sugar profile, exist-

ence of acetonemia, complete urinalysis, protidemia, lipemia, urea in the blood, liver function tests, hemogram, renal adequacy test and, in a sizable number of cases, liver biopsy puncture[12] In the hospitalized patients, a study of the carbohydrate metabolism was performed with the standard curves (Allbright's technique), intravenous glucose, intravenous insulin glucose and adrenalin insulin In six cases the action of chlorpropamide in intravenous administration was studied

Dosage Schedule for Chlorpropamide

The drug was administered in most of the cases in 2 doses (1 after breakfast and 1 after lunch) In some patients the drug was given after dinner The highest total dosage administered thus far is 71 gm , the longest duration of therapy 130 days All patients are continuing on their respective dosage schedules

Techniques and Standards Used

Addis counts were made by the usual technique Blood tests were made as follows urea, modified enzymatic technique of Wrenn, glucose, the

TABLE 2
DEFINITIONS OF STANDARDS OF CONTROL AND EVALUATION FOR CHLORPROPAMIDE

Control	Blood sugar mg % (Somogyi Nelson)	Total urine sugar gm	Acetone
Excellent	80 110	none	none
Good	111 130	To 5 per cent of the food glucides	none
Fair	131-170	To 10 20 per cent of the food glucides	none
No control	Higher levels	Higher levels	—
Efficient			
Partially efficient			
Inefficient			

Somogyi Nelson technique alkaline phosphatase, the Bodansky method blood proteins the techniques of Calvary, Cohn, Wolfson and Ichiva cholesterol, the method of Bloor, Sheftel, and Hill, Maclagan, the original technique with adaptation of electrophotometric reading, Hanger, the

test, the usual technique

Designations for standards of effectiveness of chlorpropamide in the control of diabetes in relation to blood sugar and glycosuria are shown in TABLE 2 Diabetic groupings according to response to chlorpropamide are shown in TABLE 3

BLOOD LEVELS OF CHLORPROPAMIDE IN NORMAL MEN FOLLOWING CHRONIC ADMINISTRATION

Michael Carlozzi, Domenic G Iezzoni, Lawrence Silver
Chas Pfizer & Co Inc Brooklyn N Y

As part of a large study program on the efficacy and activity of the new oral hypoglycemic agent chlorpropamide it was felt that a knowledge of the blood levels at various dosages following chronic oral administration would be of importance We were already aware that only small amounts are excreted in the urine Therefore it seemed necessary to determine whether serum levels continued to rise with repeated administration or whether some plateau value would ensue

Method

Three groups of normal male volunteers were placed on a daily dosage schedule of either 0 25 0 50 or 1 0 gm chlorpropamide daily The group

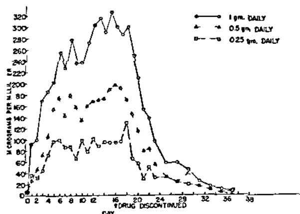


FIGURE 1 Chlorpropamide serum levels

taking the highest dose was composed of 20 men, the other 2 groups comprised 10 men each A fasting blood sample was drawn on the morning of the first day and the drug was administered orally immediately afterwards

received 100 mg daily The entire group on 1 0 gm daily had the drug

ried out in our laboratories by the method of Toolan and Wagner [1]

TABLE 1
BLOOD LEVEL OF CHLORPROPAMIDE, 0 25 GM DAILY (µg ml)

Patient	Day 1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	25	27	29	31	33	35	37
21	63	30	41	43	95	8	35	43	35	77	43	90	7	40	63	80	5	75	48	4	18	37	10	0	35	42	0	0	0	0
22	44	30	40	83	—	75	90	375	—					50	48	75	82	250	55	63	25	48	32	—	—	15	30	—	10	10
23*	55	41	4	8	125	145	120	130	85	112	103	185	158	110	160	135	207	130	125	127	90	110	80	15	77	50	40	32	25	20
24	45	49	45	8	99	100	77	80	60	75	58	100	5	—	85	93	113	—	—		—	—	—	—		—	—	—	—	—
25*	43	42	6	74	113	95	88	130	100	162	92	125	90	123	100	120	110	123	110	125	90	125	50	40	50	50	35	32	25	20
26*	46	52	52	67	110	115	127	118	70	145	98	107	93	113	105	123	123	10	102	85	70	82	68	—	50	55	45	35	—	25
27	39	43	41	3	92	110	83	92	70	85	65	50	80	90	97	98	83	82	60	47	30	50	30	85	15	30	18	20	—	10
28	39	44	68	97	113	103	90	120	40	85	90	100	90	140	123	123	125	107	102	72	38	67	48	10	18	30	—	12	0	0
29*	40	44	69	62	50	87	75	77	60	80	75	75	60	87	102	113	128	83	55	70	58	0	57	48	42	15	30	20	10	10
30*	43	40	64	73	85	92	75	100	77	77	75	0	65	98	68	102	85	0	70	48	18	62	47	—	15	40	0	0	0	0
														Mean no drug		94	96	129	66	57	28	51	30	12	23	19	16	11	3	5
Mean	46	42	54	73	98	100	86	88	66	93	8	101	88	95	95	119	131	102	92	90	63	90	60	34	37	42	30	28	33	15

* Started on 100 mg daily on sixteenth day others discontinued

TABLE 2
BLOOD LEVEL OF CHLORPROPAMIDE 0.5 GM DAILY (μg/ml)

Patient	Day 1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	25	27	29	31	33	35	37
31	15	23	48	63	93	90	85	119	98	110	93	115	110	130	150	125	115	90	90	50	30	25	0	0	0	—	0	0	0	0
32*	35	62	85	156	115	191	177	235	188	238	235	239	240	263	250	270	293	410	222	162	132	182	155	27	80	67	42	—	35	25
33*	30	63	90	122	151	190	173	220	160	185	183	200	206	195	220	250	220	180	180	152	90	125	105	62	50	35	35	—	25	20
34*	21	48	80	98	190	115	109	160	165	90	148	100	162	180	185	175	167	150	135	105	88	—	63	35	33	20	12	0	0	0
35	20	46	63	125	163	165	148	190	145	130	145	198	200	190	227	220	200	190	165	90	80	75	57	47	10	22	20	0	0	0
36	34	57	109	153	230	265	188	245	212	215	233	250	253	260	283	275	233	200	190	150	85	125	77	63	30	32	20	30	0	0
37	36	65	110	110	210	190	150	230	200	160	210	223	165	235	193	193	—	220	177	170	115	120	90	—	50	80	27	35	20	10
38*	31	49	65	84	123	125	100	127	120	112	110	102	110	—	140	145	123	112	80	90	50	75	57	42	—	30	20	50	20	15
39	30	52	78	113	226	250	157	212	240	177	170	192	205	225	187	177	220	150	115	125	90	77	57	10	30	18	—	0	0	0
40†	5	0	0	36	60	—	—	63	58	33	75	65	52	80	55	120	83	50	30	30	0	30	—	0	0	0	0	0	0	0
														Mean off drug		198	192	170	147	117	80	84	56	30	25	38	17	13	4	2
Mean	26	47	73	106	156	176	142	180	159	135	160	168	170	173	189	210	201	213	154	127	90	127	96	47	54	41	27	25	20	15

* Started on 100 mg daily on sixteenth day, others discontinued
† Excluded from mean

TABLE 3
BLOOD LEVEL OF CHLORPROPAMIDE 1 0 GM DAILY (µg ml)

Patient	Day 1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	25	27	29	31	33	35	37
1	105																							30	35	30	18	—	0	0
2	85																							93	62	72	32	27	15	10
3	92																							40	15	35	10	0	0	0
4	85																							50	45	50	30	35	10	10
5	91																							40	18	25	15	0	0	0
6	100																							68	70	62	35	27	20	10
7	128																							—	40	60	30	20	10	10
8	88																							35	30	25	40	0	10	0
9	93																							50	50	50	30	20	10	10
10	79																							38	40	42	15	12	0	0
11	75																							90	50	48	45	40	20	12
12	82																							65	47	42	18	0	0	0
13*	58																							0	40	48	25	0	0	0
14*	0																							12	12	30	17	10	0	0
15	77																							60	65	52	15	0	0	0
16	80																							65	62	35	12	0	0	0
17*	63																							20	35	60	18	0	0	0
18	98																							73	77	50	27	—	10	10
19	0																							77	153	80	35	30	20	20
20	77																							18	—	0	0	0	0	0
Mean	92																							57	59	45	24	14	8	5

* Excluded from mean al

Results and Discussion

The average daily serum levels of *chlorpropamide* for the three dosages administered are given in FIGURE 1 TABLES 1, 2, and 3 show that the individual serum levels within each group were spread over a considerable range

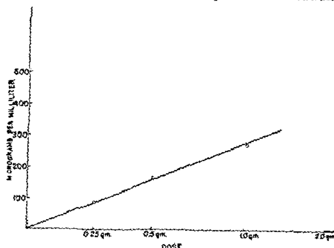


FIGURE 2 Log dose chlorpropamide serum level curve

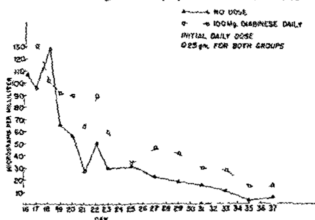


FIGURE 3 Effect of chlorpropamide—100 mg daily—on maintenance of serum level

of values However, each subject on a day to day basis, had reasonably consistent values It is apparent that at 0 25 and at 0 50 gm daily the serum levels reach a plateau in 3 to 5 days around which there is some daily variation At 1 0 gm daily it appears that the plateau is reached somewhat later there appears to be a further cumulative effect after 5 to 6 days Averages of the plateau levels for each dose yielded a straight line log-dose curve as shown in FIGURE 2 The rate of excretion of *chlorpropamide* is con-

siderably slower than that for tolbutamide and presumably accounts for

chlorpropamide levels is substantially the same in those individuals who were maintained on chlorpropamide at 100 mg daily after discontinuing the prior larger daily doses as in those where the medication was discontinued

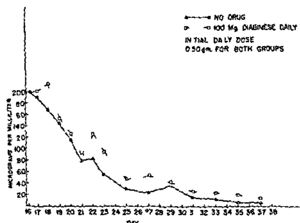


FIGURE 4

after 15 days During the course of the study there were no subjective or objective symptoms observed However many of the subjects did experience hunger and supplemented their diet with chocolate and hard candies

Summary

Various doses of chlorpropamide were given orally to normal volunteers Serum levels were followed for thirty seven days No untoward effects were observed

Reference

1 TOLAN T & R L WAGNER JR 1959 The physical properties of chlorpropamide and its determination in human serum Ann N Y Acad Sci 74 3) 449

SULFONYLUREA DRUGS IN THE TREATMENT OF DIABETES MELLITUS

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Introduction

The National Institute of Nutrition of Argentina first began clinical investigation of the sulfonylurea drugs with carbutamide in March 1956. The results were reported at a meeting of the Argentine Association of Nutrition and Dietetics in October 1956 and were published in 1957.[1] We expanded our work to include tolbutamide, and a report of the results obtained with this drug was also published.[2] Some of us have reported separately on our experiences with the sulfonylurea drugs.[3-5] Two conferences were held on sulfonylurea drugs by the Argentine Medical Association (in 1957 and 1958) and two special sessions were held by the Argentine Association of Nutrition and Dietetics.

Theoretically, the ideal sulfonylurea drug would provide the following: (1) *minimal gastrointestinal intolerances, with no acute or chronic side* reactions or toxicity and an ample margin of safety, that is, a wide difference between the therapeutic dose and the toxic dose; (2) a favorable action in the maj[illegible] regardless of age, physical condition, nutritional status, [illegible]ions, or intercurrent disease, [illegible]o the normal physiological mechanism that maintains sugar metabolism.

An analysis of all available preparations reported in the literature was published by one of us in June 1958[6] and is summarized in TABLE 1. It is evident from the ample literature of the past three years that we are far from achieving the ideal therapeutic agent. For this reason, the appearance of any hypoglycemic drug makes it mandatory that we determine how close it comes to the therapeutic ideal and establish a comparison with insulin and drugs previously evaluated in regard to efficacy, dosage, intolerance, and *possible future effect on the therapy of diabetes.*

With this point of view in mind, the National Institute of Nutrition, the Chair of Pathology, and the Nutrition Clinic, in association with other Argentine centers for the study of diabetes situated in La Plata, Mendoza, and Córdoba, carried out a study of the clinical effects of chlorpropamide. The method involved rigid conformance to the same techniques at *different* places, which invests this project with both a unity and a diversity that *affect the significance of the results.*

TABLE 1

CONSENSUS ON ACTION OF AND INDICATIONS FOR BENZENESULFONAMIDES*

Benzenesulfonamides	Probable locus or mode of action
p aminobenzenesulfamidoisopropylthiodiazole	(1) Pancreas
	(2) Inhibition of insulin degradation
p aminobenzenesulfonylbutylurea	(3) Liver enzyme systems
p methylbenzenesulfonylbutylurea	(4) Peripheral effect on carbohydrate metabolism

Criterion of case selection	Factors indicating use of benzenesulfonamides
(1) Age	(1) More than 50 years old
(2) Duration of diabetes	(2) Less than 10 years
(3) Duration of insulin treatment	(3) Less than 5 years
(4) Insulin dosage	(4) Less than 40 U
(5) Constitutional type	(5) Pyknic with obesity and essential hypertension tendencies
(6) Fasting blood sugar level before treatment	(6) Not more than 300 mg /cc
(7) Ketosis tendency	(7) None

* To June 1958

MATERIAL AND METHODS EMPLOYED

Characteristics of the Patients Studied

The drug was used on 200 patients, but data have been tabulated for only 130, these patients have been treated for a sufficient length of time to permit an evaluation to be made (FIGURE 1)

Conditions Under Which the Study Was Conducted

Form of control Of the total of 130 patients whose data have been tabulated, 30 (23 per cent) were studied while they were hospitalized, and 100 (77 per cent) were observed in the outpatient department The latter were

selected from patients who assured us that they would follow instructions carefully To evaluate fully the effect of the drug we administered it to patients having various types of diabetes After observations of responses were made, we continued our investigation of those individuals who had some degree of satisfactory response to the drug

The diet followed A nutritional study was conducted in order to acquire a knowledge of the diet of all patients A dietary interview was held with each patient during which a detailed history of his dietary habits was recorded

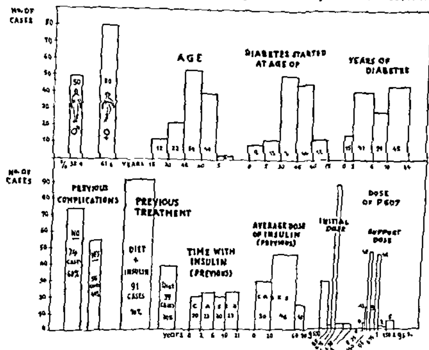


FIGURE 1 Characteristics of 130 cases under observation

Dietary regimens, with the characteristics of these normal regimens introduced, were designed to provide each patient with the caloric value appropriate for his or her nutritional status and to include approximately 200 gm of carbohydrates, this amount was reduced in certain cases and increased in others to as much as 300 gm It is understood, of course, that the other values— those of the minerals and vitamins—were the same as in the normal regimen in every case

Examinations and tests used to evaluate the response to therapy The observation of each patient in the study was carried out under strict clinical, medical, and laboratory control Clinical determinations included body weight, diuresis and any subjective or objective symptoms that appeared Laboratory determinations included blood sugar before breakfast, blood sugar profile during the day, total daily urine sugar and urine sugar profile, exist-

ence of acetonemia, complete urinalysis, protidemia, lipemia urea in the blood, liver function tests, hemogram, renal adequacy test and, in a sizable number of cases, liver biopsy puncture [12] In the hospitalized patients, a study of the carbohydrate metabolism was performed with the standard curves (Allbright s technique), intravenous glucose, intravenous insulin glu cose, and adrenalin insulin In six cases the action of chlorpropamide in intravenous administration was studied

Dosage Schedule for Chlorpropamide

The drug was administered in most of the cases in 2 doses (1 after breakfast and 1 after lunch) In some patients the drug was given after dinner The highest total dosage administered thus far is 71 gm , the longest duration of therapy, 130 days All patients are continuing on their respective dosage schedules

Techniques and Standards Used

Addis counts were made by the usual technique Blood tests were made as follows urea modified enzymatic technique of Wrenn glucose the

TABLE 2
DEFINITIONS OF STANDARDS OF CONTROL AND EVALUATION FOR CHLORPROPAMIDE

Control	Blood sugar mg % (Somogyi Nelson)	Total urine sugar gm	Acetone
Excellent	80–110	none	none
Good	111 130	To 5 per cent of the food glucides	none
Fair	131 170	To 10–20 per cent of the food glucides	none
No control	Higher levels	Higher levels	—
Efficient	Produces excellent or good control		
Partially efficient	Produces poor control Reduces insulin use no less than 50 per cent Has stabilizing action on glucose homeostasis		
Inefficient	Apparently no action on diabetes		

Somogyi Nelson technique, alkaline phosphatase, the Bodansky method blood proteins, the techniques of Calvary, Cohn, Wolfson and Ichiva cholesterol, the method of Bloor, Sheftel, and Hill, Maclagan, the original technique with adaptation of electrophotometric reading, Hanger, the original technique, including maintaining in the dark for 48 hours Kunkel, the original technique, blood bilirubin the technique of Mallory and Evelyn, erythrosedimentation, the technique of Westergren, and the cytological test, the usual technique

Designations for standards of effectiveness of chlorpropamide in the con trol of diabetes in relation to blood sugar and glycosuria are shown in TABLE 2 Diabetic groupings according to response to chlorpropamide are shown in TABLE 3

TABLE 3
CLASSIFICATION OF DIABETICS

Group	Definition
1	Controllable with proper diet and insulin of as much as 20 U. Insulin withdrawal does not produce ketosis
2	Controllable with proper diet and insulin dosage of as much as 59 U. Insulin total withdrawal (even if less than 20 U) produces ketosis. History of ketosis exists
3	Controllable with proper diet and more than 60 U. of insulin. With or without past history of coma

RESULTS

General

The effects of chlorpropamide on blood sugar levels in normal individuals are shown in FIGURE 2. FIGURE 3 shows the effect on blood sugar levels of diabetics and FIGURE 4 the effect on the control of diabetes as indicated by blood sugar levels and glycosuria.

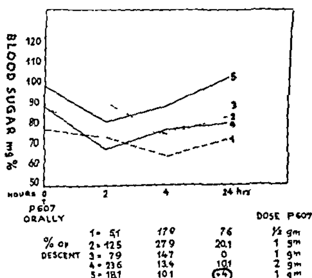


FIGURE 2. Glycemia response with chlorpropamide in five normal subjects.

Special

The complementary action of chlorpropamide on the control of cases of diabetes treated with insulin is illustrated in FIGURES 5 to 8. FIGURE 6 shows the control exercised by tolbutamide and chlorpropamide in diabetics undergoing surgery. FIGURE 7 shows the action of chlorpropamide in two

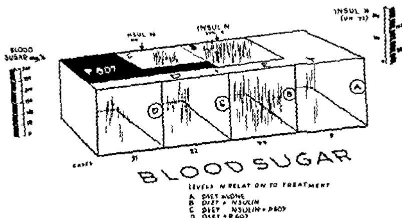


FIGURE 3   Blood sugar levels in relation to treatment

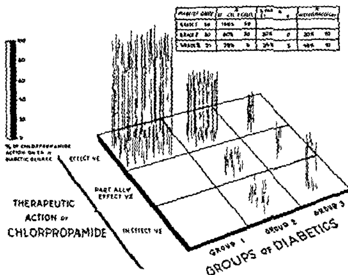


FIGURE 4   Results in 130 diabetic patients treated with adequate diet and chlorpropamide for 4 months

cases of pancreatic disease—one chronic pancreatitis with steatorrhea and diabetes the other with acute pancreatitis, operated for obstructive hepatitis (cholecystojejunostomy) three years before the beginning of diabetes The action of chlorpropamide in one case of hyperthyroidism in diabetes is shown in FIGURE 8 Separate observations were made of the effect on blood sugar levels of methylthiouracil, methylmercaptoimidazole, carbutamide, chlorpropamide, and chlorpropamide with methylthiouracil In one case of

FIGURE 5 Complementary action of chlorpropamide in insulin treated diabetics

diabetes and vitiligo good control was possible with chlorpropamide (FIGURE 9) The effect on blood sugar level of chlorpropamide administered intravenously is shown in FIGURE 10

No beneficial effect was observed from chlorpropamide administered to two cases of moderate to intense acidosis Three cases of mild ketosis were correctable with the drug

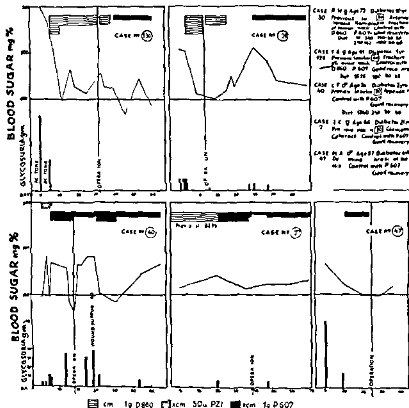
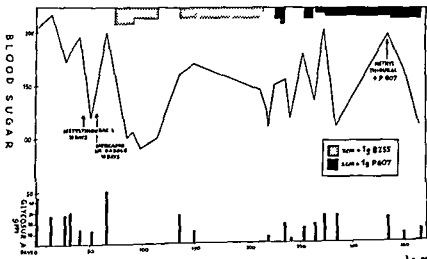


FIGURE 6 Surgical cases treated with tolbutamide and chlorpropamide

During treatment with chlorpropamide relatively high blood sugar levels were observed without glycosuria or with only slight glycosuria This apparent elevation of the renal threshold is accompanied by a decrease in diuresis

In two cases we have observed a relationship between dosage and degree

FIGURE 7 Action of chlorpropamide on two cases of pancreatitis



BLOOD SUGAR mg%

xcm = 1g BZ55

xcm = 1g P607

GLYCOSURIA gms

DAYS 0 10 20 30 40 50 60 70 80

CASE Nº 42 A.P 16yr diabetes vitiligo age 60yr
♂ BZ55 2gm P607 1g

FIGURE 9 Action in relation to dosage

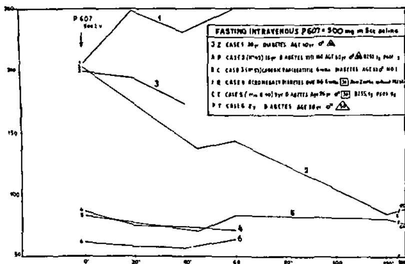


FIGURE 10 Effect on blood sugar level of intravenous administration of chlorpropamide

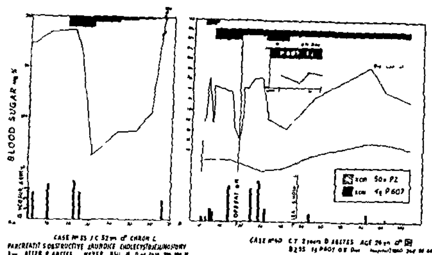


FIGURE 11 Action of chlorpropamide in relation to body weight and dosage

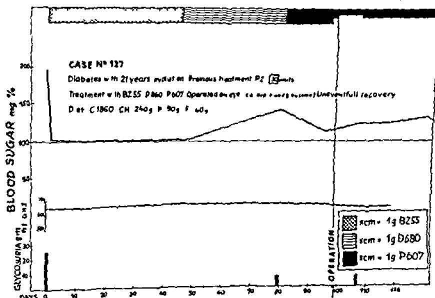


FIGURE 12 Action of drugs in patient whose body weight was kept constant

of the drug used and the degree of control in a patient in whom the body weight was maintained at a constant level (FIGURE 12)

Side Effects in the Administration of Chlorpropamide

There was 1 gastrointestinal case (constituting less than 1 per cent), and no renal or hepatic cases Hematic effects were limited to 2 cases of leuko-

penia (1 6 per cent) and cutaneous effects to 15 cases (11 5 per cent), 13 of flushing (10 per cent) and 2 of pruritus with exanthema (1 6 per cent) The flushing observed appeared similar to that produced by the ingestion of nicotinic acid In two cases where the flushing was intense, it was related

(0 8 per cent) of cyanosis that disappeared spontaneously and did not reappear when dosage was decreased from 0 5 gm to 0 25 gm of chlor propamide There were
presented symptoms of
reducing the dosage of chl
symptoms of a mild hypoglycemia, but these cases were transitory and could not be classified definitely as actual hypoglycemic episodes In one case (patient 26) epigastric and precordial discomfort was experienced 4 hours after ingestion of 0 5 gm of chlorpropamide, this coincided with a clearly low blood sugar

Side effects were related to dosage They appeared when the dosage was 1 gm daily or higher, and rarely with dosage of 0 5 gm daily (case 89, cyanosis) We have the impression that the margin between the therapeutic dosage and the dosage that produces side effects is small in other words, the margin of safety is narrow

Discussion and Conclusions

Classification of Diabetes

From our experience with sulfonylurea drugs and, particularly, with chlorpropamide in the treatment of diabetes, it is possible to subdivide diabetics into three groups Our classification is based on symptoms that are obvious and reliable After studying all symptoms that can be used for classification, it was decided that the most constant, in relation to the results obtained, are ketosis, diabetic coma, and previous insulin dosage required before establishing therapy with the sulfonylurea drugs On the basis of the patients' responses with respect to these three factors, we have defined

TABLE 3

TABLE 4 shows the control achieved with other sulfonylurea drugs as observed in earlier studies carried out with the same methods used with chlorpropamide

Administration of the Drug

The minimum effective maintenance dosage of chlorpropamide was 0 125 gm (in 4 cases) to 0 5 gm per day, the latter was the most commonly used dosage One-half gram daily was effective in 38 cases To be effective, it

was necessary to increase dosage to 1 gm daily in another 33 cases When it was necessary to increase the maintenance dosage further, 1 5 gm was effective in 2 cases out of 3, and partially effective in 1 other In 7 cases in which the drug was ineffective, the dose was increased to 2 gm , with some improvement With chlorpropamide, the glucose-regulating effect is sometimes delayed for as long as 1 week or 10 days, during which time the patient should be observed carefully to determine if a change in dosage is necessary

TABLE 4
RESULTS WITH CARBUTAMIDE OR TOLBUTAMIDE IN 130 DIABETICS

Diabetic group	Carbutamide					
	Effective		Partially effective		Ineffective	
	Percent ages	No of cases	Percent ages	No of cases	Percent ages	No of cases
1	73 80	31	9 52	4	16 66	7
2	68 05	49	15 27	11	16 66	12
3	18 75	3		0	81 25	13
	Tolbutamide					
1	61 42	43	17 14	12	21 42	15
2	48 07	25	34 61	18	17 30	9
3	37 50	3	25 00	2	37 50	3

Mode of Action

The investigation as conducted does not give any clue to the manner in which chlorpropamide acts Its action appears to be similar to that of other sulfonylurea drugs although effective dosage with chlorpropamide is lower ... dosages of the three ... the dosages of

Our investigations do not determine definitely all possible effects of long term therapy

We have used chlorpropamide for five months, a rather short time, therefore we can reach only very preliminary conclusions

Tolerance of Chlorpropamide

The tolerance of effective dosage has been good as the percentages of intolerance and of side effects indicate Gastrointestinal intolerance increases with dosages greater than 1 gm daily

Usefulness of Chlorpropamide

Treatment with Chlorpropamide and the Importance of Dietary Measures

Regardless of the group in which a diabetic is classified a basic factor in the treatment is a balanced diet that normalizes body weight It has been

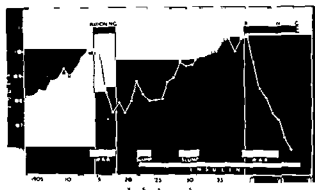


FIGURE 13 Comparative mortality indexes for diabetic patients in England and Wales

observed that with chlorpropamide as with other sulfonylurea drugs stability is endangered when the patient s weight increases Dramatic evi lence of ho h h

the mortality rate for diabetics dropped to a marked degree No more graphic evidence of the importance of adhering strictly to dietary regimens exists

SUMMARY

Chlorpropamide was studied at the National Institute of Nutrition with the collaboration of diabetes centers of La Plata Córdoba and Mendoza The medication was administered to a total of 200 diabetic patients all more than 15 years of age Thus far however detailed tabulations have been completed on only 130 of these cases Patients were subdivided into 3 classifications

Group 1 those controlled with insulin, dosage of as much as 20 U, but in whom withdrawal of insulin did not produce acidosis

Group 2 including (a) those controlled with less than 20 U of insulin, but in whom acidosis appeared on withdrawal, (b) those requiring between 21 and 59 U of insulin and (c) those presenting a history of acidosis

Group 3 those requiring more than 60 U of insulin or having a history of diabetic coma Using this classification of diabetic disease the results shown in TABLE 5 were observed with chlorpropamide in 130 patients of this series

TABLE 5
RESULTS WITH CHLORPROPAMIDE IN 130 DIABETIC PATIENTS

Diabetic group	Total cases	Effective		Partially effective		Ineffective	
		Percentages	No of cases	Percentages	No of cases	Percentages	No of cases
1	59	100	59	—	—	—	—
2	50	60	30	20	10	20	10
3	21	28	6	24	5	48	10

Hypoglycemic action has been demonstrated with chlorpropamide in normal individuals As for comparing its efficacy with carbutamide and tolbutamide equal hypoglycemic action was obtained with smaller doses of chlorpropamide Otherwise, the same factors appear to govern the effectiveness of these drugs Daily dosage ranged from 0 1 to 2 0 gm most frequently an initial dose of 1 0 gm (67 per cent of the cases), with 0 5 to ... Side effects included

Tests of hepatic and renal function revealed no disturbance, even in those patients receiving the drug for as long as 130 days The action of chlorpropamide has been studied in relation to a diet that contains less carbohydrate than a normal diet

ACKNOWLEDGMENTS

We express our appreciation of the collaboration of Boris Rothman Vicente Osvaldo Perrone Enrique Benito De Marco Mario Calvagno,

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PRELIMINARY CLINICAL AND EXPERIMENTAL STUDIES WITH CHLORPROPAMIDE IN DIABETES MELLITUS

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The purpose of this communication is to describe our experiences in the clinical management of sixty diabetic patients who have been treated with chlorpropamide for periods varying from a few weeks to six months. In addition the results of some comparative experiments on the action of tolbutamide and chlorpropamide *in vitro* are given These suggest that the mode of action of the two drugs upon hepatic enzyme systems is similar but there seems to be a quantitative difference between them

Clinical observations have been made on sixty adult diabetic patients twenty one males and thirty nine females In view of earlier experiences

been made from patients attending the routine practice of the diabetic service of this hospital

In contrast with earlier clinical trials conducted in the Diabetic and Metabolic Unit of the Alfred Hospital [1,2] many of the patients in this series were not admitted to hospital for continuous observation It was felt that experience gained during the past three years was sufficient to enable us to assess the efficacy of chlorpropamide without the need for hospitalization In consequence a number of patients were instructed to present the results of urine tests taken at regular times each day and spot blood sugar estimations

gations were undertaken for evidence of hepatic ... abnormalities Patients transferred from maintenance doses of insulin were instructed to report daily either in person or by telephone and to present the results of urinalyses performed four times each day

Almost without exception the loading dosage prescribed was 1 0 gm per day and the maintenance dose 0 5 gm per day given in divided doses of 0 25 gm morning and evening In any case that failed to respond the ... few instances to ... day for a short ... ients considered ... atment and the number failing to do so The relatively large number not responding within the first 4 weeks of treatment can be related to several such factors as inappropriate election of patients and attempts to transfer cases from insulin therapy TABLE 1 shows the distribution of the patients by age and sex

and TABLE 2 shows the over all results obtained in the entire series It is convenient to present the results obtained for 4 categories of patients (1) previously untreated, (2) previously treated by dietary restriction, (3) previously treated by diet and tolbutamide, and (4) previously treated by diet and insulin

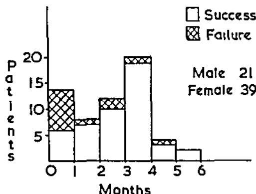


FIGURE 1

TABLE 1
DISTRIBUTION OF PATIENTS BY AGE AND SEX

Age	Male	Female
20–30		1
30–40		1
40–50	2	1
50–60	4	5
60–70	9	15
70–80	4	13
80–—	2	4
	21	39

(1) Fourteen patients previously untreated were placed upon an appropriate restriction of diet with chlorpropamide in an initial dosage of 1 0 gm and a maintenance of 0 5 gm per day A number of these were assessed as likely to require insulin for control Twelve patients responded satisfactorily, while 2 failed to do so

(2) Twenty patients who had shown inadequate or intermittently satisfactory response to dietary restriction for periods from a few months to several years were treated along similar lines Seventeen are considered to have shown a satisfactory response to chlorpropamide therapy, while 3 failed to do so

(3) Four patients who had been treated for periods from 1 to 4 months

with dietary restriction and with tolbutamide in conventional dosages of 1.5 gm. per day and all of whom had failed to show satisfactory response to this drug, were transferred to chlorpropamide in dosages of 0.25 gm. twice daily. Three showed a satisfactory control while 1 failed to respond.

(4) Twenty-two patients whose condition had been regarded as controlled more or less satisfactorily by dietary restriction together with varying dosages of insulin were transferred to chlorpropamide therapy. Successful control was established in 15, while 7 failed to respond.

Chlorpropamide was used intermittently for periods of a few days to 1 to 2 weeks in treating still another group of 12 patients who, for various reasons, had become unstable while on dietary restriction alone. It was felt that a short trial of a sulfonyl drug might restore control and that the drug might

TABLE 2
RESULTS OBTAINED IN ENTIRE SERIES

Previous treatment		Success	Failure
Nil	14	12	2
Diet alone	20	17	3
Diet plus tolbutamide	4	3	1
Diet plus insulin	22	15	7
	60	47	13

then be discontinued. In every instance the results obtained were considered entirely satisfactory as after a short course of dosages of 0.25 to 0.5 gm. per day it was possible to discontinue its use, the patients being restored to their previous state of equilibrium.

TABLE 3 gives some details of the 13 patients who failed to respond. Two patients previously untreated, one a male of 54 and the other a female of 64, both with a history of diabetes of short duration who on general principles were considered likely to respond favorably, showed no evidence of control on dosages of 1.5 gm. and 1.0 gm., respectively, although these dosages were continued for more than a fortnight. In neither case was there any evidence of an appreciable fall in the level of blood sugar.

In three instances, patients whose dietary control had been regarded as unsatisfactory for several months were tried with chlorpropamide therapy.

These patients were a female of

were treated for 6 to 8 weeks with

these patients was subsequently

satisfactorily controlled wi[illegible]

Seven of the 22 patients previously taking insulin failed to show satisfactory control when transferred to chlorpropamide. In 5 of these the duration of diabetes ranged from 6 to 10 years and their daily requirement of insulin varied between 20 and 50 U. per day. In two instances, a female of 36 and

another female of 24, chlorpropamide was used because of the brittle nature of the diabetes, and in the hope that it might facilitate the action of insulin In both instances it failed in less than 1 week, 1 patient developing a severe ketosis that necessitated admission to hospital

Four patients who had been inadequately controlled by diet and tolbutamide were transferred to chlorpropamide One patient failed to respond She was 80 years old and, earlier in her diabetic history, had been controlled with 56 U of protamine zinc insulin each day She had been transferred to 1 5 gm tolbutamide per day, but showed no response and, likewise, was not influenced by chlorpropamide

In this trial there have been no unexpected episodes either in response or in failure to respond to this new sulfonyl compound The side effects observed

TABLE 3
DATA FOR THIRTEEN PATIENTS FAILING TO RESPOND TO TREATMENT

	Sex	Age	Wt	Dose	Duration	Side effects	Duration of diabetes
Previously untreated	M	54	158	1 5	2 weeks	Nausea	1 year
	F	64	155	1 0	2 weeks	—	4 months
Diet alone	F	73	124	0 75	3 months	—	4 months
	M	65	145	1 5	1 month	Nausea	2 months
	F	65	143	0 75	2 months	—	6 years
Diet and tolbutamide	F	80	200	0 50	5 months	—	5 years
Diet and insulin	F	63	177	1 0	2 weeks	—	12 yrs 40 NPH
	F	24	146	1 0	2 weeks	Ketosis	9 yrs 20 NPH
	F	52	136	1 0	2 weeks	—	6 yrs 24 NPH
	F	59	143	1 0	2 weeks	—	10 yrs 24 R 24 PZ
	F	74	86	1 0	1 week	Insulin allergy nausea	20 yrs 40 NPH
	F	72	150	0 5	2 weeks	Diarrhea	2 yrs 28 R 20 PZ
	F	36	136	0 75	1 week	Ketosis	20 yrs 48 NPH

have been relatively insignificant A small number of patients complained of nausea and this was more apparent in those cases in which the dosage was raised to more than 0 5 or 0 75 gm per day In fact almost every case in which the dose was raised to more than 0 75 gm daily there was some complaint of nausea or dizziness In 2 instances a photosensitive rash, no different from that observed in other forms of sulfonamide therapy occurred and, in both this was of minor degree On cessation of the drug the rash soon disappeared In 2 instances albuminuria developed and the drug was discontinued despite the fact that there was no significant alteration in renal efficiency tests It did not seem that any obvious renal damage had occurred In all instances in which hepatic efficiency tests were performed there was neither clinical nor biochemical evidence that any hepatic disturbance was

occurring or had occurred as a result of chlorpropamide therapy In no instance was any depression of blood formation observed In three cases a curious effect was noted by patients after the consumption of preprandial alcoholic drinks, all complained of sensations of flushing of the face, which was obvious to onlookers and lasted for several hours

In view of our previous experience that carbutamide and tolbutamide both apparently inhibit liver transaminase it was decided to investigate the effect of chlorpropamide and tolbutamide on the oxidation of C^{14} labeled amino acids by liver slices, using the drugs at a more physiological concentration than in previous enzyme experiments[3]

The slices were incubated in Krebs phosphate buffer at a glucose concentration of 100 mg per 100 ml and a C^{14} DL alanine concentration of 40 mg

TABLE 4
EFFECT OF TOLBUTAMIDE AND CHLORPROPAMIDE ON THE OXIDATION OF C^{14} ALANINE BY LIVER SLICES

	No Series	$C^{14}O_2$ recovered as cpm/mg liver/hour	Difference	P
Experiment 1				
Control	6	108 5 ± 9 8 (S E M)		
Chlorpropamide	6	154 7 ± 14 2	46 2	<0 01
Experiment 2				
Control	6	98 2 ± 3 4		
Tolbutamide	6	106 3 ± 3 4	8 1	<0 01

per 100 ml Either chlorpropamide or tolbutamide was added to 1 of each pair of slices to give a final concentration of 30 mg per 100 ml

The $C^{14}O_2$ was collected on a KOH soaked paper spill in the center well of the Warburg vessel used, then eluted and precipitated as $BaC^{14}O_3$ the radioactivity counted was corrected to infinite thinness The results of this study are shown in TABLE 4 It is seen that both compounds accelerated the oxidation of the keto acid moiety of the amino acid in contradistinction to the result that might have been anticipated from the enzyme experiment

Although these results appear at variance it is likely that the apparent inhibition of transaminase may well have been due to accelerated oxidation of the acceptor α ketoglutaric acid in the enzyme experiment, leading to a diminution in the amount of glutaric acid recovered However, the suggestion that at least part of the action of these compounds is due to inhibition of neoglucogenesis is still valid, although the effect may be an acceleration of oxidation of Krebs cycle intermediates rather than an inhibition of their formation It is interesting to note that at the same concentration chlorpropamide is roughly five times as effective in promoting the oxidation of alanine as is tolbutamide

To practicing physicians, the discovery of the hypoglycemic action of

the sulfonylureas has provided a fresh interest in the management of diabetic

of diabetic patients However it has been obvious for some time that control of hyperglycemia does not prevent patients from developing peculiar complications that affect the vascular and nervous systems This has led to the speculation that diabetes mellitus is not always a problem of insulin deficiency alone and that substitution therapy with insulin does not prevent the development of complications at least in some patients It is possible that physicians tend to forget that insulin has effects on the metabolism of proteins and of fats impossible to assess by any simple clinical means and that they place too much emphasis on easily performed tests for glucose in blood and urine

Consequently when a chemical substance whose action is as yet unknown is found to be capable of reducing hyperglycemia in some patients and not in others it is not surprising that something resembling a therapeutic tumult has occurred Eighty years ago Lancereaux[5] pointed out the clinical difference between *diabete maigre* and *diabete gras* a distinction emphasized by Himsworth[6] in 1949 and proved by Lawrence and Bornstein in 1950[7] It seems that the sulfonylurea compounds are capable only of influencing those patients who are still able to produce some insulin of their own naturally fears have arisen that they may be disturbing bodily functions in a harmful fashion Certainly it seems possible from the *in vitro* studies already

these compounds alter or even correct such a deviation from normal metabolism This could offer an explanation for those cases whose hyperglycemia previously controlled with appreciable dosages of insulin is capable of control with small amounts of these new drugs given each day

From this brief experience it would seem that chlorpropamide is capable of controlling the hyperglycemia of an appreciable number of diabetic patients who have developed the complaint in middle age

No harmful side effects have been observed thus far in any of the patients treated with chlorpropamide

There appears to be no qualitative difference between the patients who respond to carbutamide and those responsive to tolbutamide or chlorpropamide

There appears to be a significant difference between the required dosages of chlorpropamide and tolbutamide in the patients who show response to these drugs This is also shown by *in vitro* studies on the effect of these drugs in the promotion of oxidation of alanine

The suggestion is made that the action of these sulfonyl drugs is concerned with an acceleration of oxidation of Krebs cycle intermediates and that an inhibition of neoglucogenesis might explain their influence on some types of human diabetes

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To practicing physicians the discovery of the hypoglycemic action of

the sulfonylureas has provided a fresh interest in the management of diabetic patients For many years it has been thought desirable, if not essential, to control hyperglycemia and to reduce glycosuria The application of these principles has produced a remarkable improvement in the life and welfare of diabetic patients However, it has been obvious for some time that

deficiency alone and that substitution therapy with insulin does not prevent the development of complications, at least in some patients It is possible that physicians tend to forget that insulin has effects on the metabolism of proteins and of fats impossible to assess by any simple clinical means and that they place too much emphasis on easily performed tests for glucose in blood and urine

Consequently when a chemical substance whose action is as yet unknown is found to be capable of reducing hyperglycemia in some patients and not in others it is not surprising that something resembling a therapeutic tumult has occurred Eighty years ago Lancereaux[5] pointed out the clinical difference between *diabete maigre* and *diabete gras* a distinction emphasized by Himsworth[6] in 1949 and proved by Lawrence and Bornstein in 1950[7] It seems that the sulfonylurea compounds are capable only of influencing those patients who are still able to produce some insulin of their own naturally fears have arisen that they may be disturbing bodily functions in a harmful fashion Certainly it seems possible from the *in vitro* studies already described that some deviation of normal metabolic processes occurs although thus far we have no evidence that this is either harmful or dangerous If the diabetes of middle life is due to excessive neoglucogenesis it could be that these compounds alter or even correct such a deviation from normal metabolism This could offer an explanation for those cases whose hyperglycemia previously controlled with appreciable dosages of insulin is capable of control with small amounts of these new drugs given each day

From this brief experience it would seem that chlorpropamide is capable of controlling the hyperglycemia of an appreciable number of diabetic patients who have developed the complaint in middle age

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THE EFFECT OF CHLORPROPAMIDE ON ALLOXAN DIABETES IN MICE*

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Diabetes has been produced in mice by Kirschbaum and Bell[1] after subcutaneous injection of alloxan at the rate of 200 mg./kg. body weight, and by Lazarow[2] after intravenous administration of 50 to 70 mg./kg. More recently we have reported[3] the differences in the diabetogenic effect of intravenously injected alloxan on different strains of mice. The present paper describes some preliminary observations about the effect of chlorpropamide† on the alloxan diabetic mice.

Methods

Alloxan monohydrate (50 to 70 mg./kg. body weight) was injected into the tail vein of adult male $ZCeF_1$ hybrid mice as previously described.[3] Blood samples taken by cutting the tip of the tail were analyzed for blood [illegible]

weeks thereafter, eight of the surviving animals were normoglycemic, while the other ten were still severely diabetic.

Chlorpropamide was given by stomach tube once a day at a dosage of [illegible]

with alloxan was used as control and treated with chlorpropamide simultaneously and in the same way.

Results

The results are summarized in TABLE 1, which gives the mean blood sugar values (and standard error of the means) for the different groups of animals. The blood sugar values of the control animals before chlorpropamide was given are in the same range as those reported by Lazarow.[2] The blood sugar values of these animals did not change after the administration of chlorpropamide for 3 days (column 6, TABLE 1). A small decrease of 27 mg./100 ml. was observed after 10 days of administration (column 7, TABLE 1), but this decrease proved to be not significant ($t = 1.45$; $p > 0.1$).

Similar response was observed in the group of animals with transient diabetes. The mean blood sugar value 10 days after chlorpropamide was 27 mg./100 ml. lower, but this decrease was statistically not significant ($t = 1.1$; $p > 0.1$).

* The study reported in this paper was aided by grants from the Minnesota Division of the American Cancer Society, Inc., New York, N. Y., and the Public Health Service, Bethesda, Md.

† Supplied by Chas. Pfizer & Co., Inc., Brooklyn, N. Y.

In both groups there were animals that showed very marked decrease in blood sugar, 1 animal in the control group had a decrease of 93 mg /100 ml 10 days after administration of the chlorpropamide and 1 other, in the transient diabetes group, had a decrease of 135 mg /100 ml

In the diabetic group the administration of chlorpropamide produced a very marked decrease of the blood sugar 3 days after the beginning of treatment (column 6, TABLE 1) The mean decrease was 178 mg /100 ml, or

TABLE 1
EFFECT OF CHLORPROPAMIDE IN MICE*

Group	Number of animals	Mean blood sugar value and S E of mean†						
		5/29	6/5	6/26	6/30	7/7	7/14	7/21
Transient diabetes	8	314 ±45	401 +82	159 ±12	163 ±10	132 ±21	159 ±14	175 ±4
Permanent diabetes	10	383 ±36	561 ±52	334 ±11	156 ±19	165 ±4	183 ±7	234 ±24
Control	8	—	—	162 ±5	166 ±10	135 ±15	168 ±6	180 ±6

* Alloxan was injected on May 14 Administration of chlorpropamide (100 mg /kg) began on June 27 and continued for 16 days through July 12

† Upper figure of figure pair is mean blood sugar value in milligrams per 100 ml lower figure is standard error of the mean

All the animals had hypo

Comment

The results indicate a very marked hypoglycemic effect of chlorpropamide in the alloxan diabetic mice, which contrasts with the very slight effect observed in the control animal, and in those that were injected with alloxan but that were nondiabetic at the time of the administration of chlorpropamide These observations are at variance with other observations regarding the effect of the sulfonylureas on the alloxan diabetic animal[5,7] It would appear that chlorpropamide is more effective in decreasing the blood sugar of the diabetic mouse than in producing hypoglycemia in the normal animal

Summary

Twenty eight mice were injected intravenously with 50 to 70 mg of alloxan All the animals developed marked hyperglycemia Four weeks after injection of alloxan, 8 of the animals were normoglycemic Chlorpropamide was given to these animals by stomach tube at a dosage of 100 mg /kg of

body weight as well as to 10 others that were still diabetic, and to 8 normal controls Three days after the administration of chlorpropamide, the alloxan diabetic mice showed a very marked decrease of their blood sugar level No significant effect was produced in the other two groups of animals

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THE EFFECTS OF TOLBUTAMIDE AND CHLORPROPAMIDE ON PATIENTS EXHIBITING JAUNDICE AS A RESULT OF PREVIOUS CHLORPROPAMIDE THERAPY

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undesirable side effects that occur. Jaundice as a complication of carbutamide therapy has been reported.[1,2] Recently jaundice has also been reported[3] as a complication in tolbutamide therapy. Chlorpropamide is closely related to the above compounds, differing from them in having a chloride group substituted on the benzene ring and a propyl instead of a butyl group at the end of the chain. With such a compound, one might anticipate the same type of allergic reactions as have occurred with carbutamide and tolbutamide. In studying twenty diabetic patients for the hypoglycemic effects of chlorpropamide, three cases of jaundice with associated nausea and vomiting and with an erythematous maculopapular rash were encountered. The following case reports are illustrative.

Case 1. D. J., a 58 year old Negro female, has had known diabetes mellitus since 1949 and was controlled on diet only until February 1958. Because of glycosuria, an elevated fasting blood sugar, and weight loss, therapy was begun with 10 U. of NPH insulin each morning before breakfast. Upon this regimen her diabetes became well controlled. On March 19, 1958 insulin was discontinued and chlorpropamide therapy was instituted with a dose of 0.5 gm. twice daily. The day before, a complete blood count was normal with no eosinophilia present. Urinalysis was negative except for a 1+ sugar. Liver function tests were normal. The thymol turbidity test was 3.3 U., the alkaline phosphatase test was 3.78 Bodansky units, and the total serum bilirubin was 0.28 mg. per cent with a direct bilirubin of 0.13 mg. per cent.

Initially, the patient tolerated the medication satisfactorily and was managed with the same degree of control as with the previous insulin regimen. During the third week of chlorpropamide therapy she developed anorexia, nausea and vomiting, a dark urine, pruritus, and a skin rash. She stopped her medication on April 12, 1958. She was seen in the diabetes clinic on April 18 with the above complaints and was admitted to the Medical Service at Grady Memorial Hospital. Careful questioning revealed no history of exposure to rats or hepatotoxic agents, or to anyone with hepatitis. Needle punctures were limited to her own insulin syringe and needle, used by no one else, and to those done in our laboratory with careful precautions.

Upon examination the patient a
103° orally, the pulse rate was 116
generalized maculopapular rash

were no mucous membrane lesions, the sclerae were icteric, and there were grade 2 arteriosclerotic changes of the fundi The lungs were clear The heart was slightly enlarged to the left a grade 2 apical systolic murmur was recorded The abdominal examination revealed no evidence of hepato megaly No organs or masses were palpated The remainder of the examination was noncontributory

When the patient was admitted the blood count showed a Hb of 12 4 gm, Hct 41 and WBC 3900 with 14 per cent bands 57 per cent neutrophils 6 per cent eosinophils 17 per cent lymphocytes, and 6 per cent monocytes Platelets were adequate By April 30 the Hb had dropped to 10 5 gm and the Hct to 32 5 WBC was 9400 with 13 per cent eosinophils 46 per cent neutrophils 2 per cent bands 6 per cent metamyelocytes 31 per cent lympho cytes and 2 per cent monocytes By May 27 the blood picture had returned to normal revealing a Hb of 11 8 gm Hct 35 and WBC 6700 with 4 per cent eosinophils 56 per cent neutrophils 4 per cent bands and 36 per cent lymphocytes

Urinalysis on admission of the patient revealed a trace of sugar 4+ ace tone and 4+ bile The CO_2 combining power was 23 9 mmoles/l The 4+ acetone was thought to have been determined on the basis of starvation Liver function tests on April 18 1958 revealed a total protein of 7 1 gm albumin 3 9 gm and globulin 3 2 gm with an albumin globulin (A G) ratio of 1 22 The thymol turbidity test was 2 25 U and the cephalin flocculation test was 1+ The alkaline phosphatase test was markedly elevated to 26 9 Bodansky units The serum bilirubin was elevated to 10 7 mg per cent with a direct bilirubin of 7 65 mg per cent The prothrombin time of the patient was 43 4 sec with a control value of 11 4 sec at 100 per cent 19 8 sec at 30 per cent and 26 sec at 20 per cent

After the intravenous administration of 50 mg of vitamin K_1 the pro thrombin time of the patient on April 19 was 14 0 sec with a control value of 12 4 sec at 100 per cent 21 sec at 30 per cent and 27 6 sec at 20 per cent On April 21 the prothrombin time was 15 0 sec with the same control values as on April 19 On April 28 it was 13 0 sec with a control value of 13 4 sec at 100 per cent 21 0 sec at 30 per cent and 29 0 sec at 20 per cent Serum electrophoresis on April 25 revealed a non pecific elevation of the serum globulins

By April 24 the alkaline phosphatase had dropped to 16 1 Bodansky units After the patient was discharged from the ho pital the alkaline pho phatase was 5 66 Bodansky units on May 20 4 30 on May 27 6 88 on June 3 and 3 84 on June 10 1958 Since the last date the e value have remained normal

On April 21 the total serum bilirubin was 8 5 mg per cent with a direct bilirubin of 6 1 mg per cent On April 22 the total serum bilirubin was 10 9 mg per cent with a direct bilirubin of 7 9 mg per cent On April 28 total serum bilirubin was 6 3 mg per cent with a direct bilirubin of 5 0 mg per cent After the patient was di charged from the hospital on May 20 the total serum bilirubin was 2 45 mg per cent with a direct bilirubin of 1 6 mg per cent On May 27 the total serum bilirubin was 1 5 mg per cent

per cent On June 3 the total serum
^ per cent }
range

While the patient was in the hospital and si ce al proteins albumin globulin ratios cephalin flocculation tests and thymol turbidity tests have been within normal limits

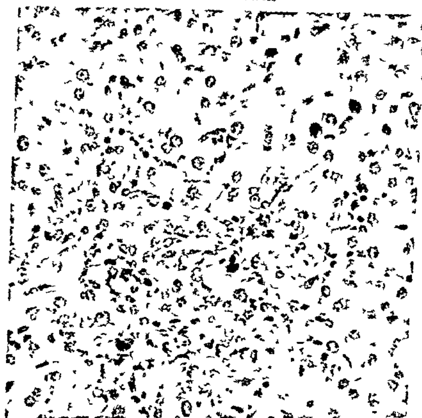


FIGURE 1 Hepatocellular damage as revealed by biopsy × 160

nsy was *obtained* on April 22 1958 FIGURE 1 shows the micro here was a marked increase in mitotic activity th pattern Hepato cellu re present Kar eosinophilic granulocyte smaller canalicul was present The changes were those of and hepatocellular damage A chest X ray was suggestive of slight cardiomegaly but was otherwise negative

For the first 4 days in the hospital a daily temperature elevation as high as 102 2° to 103 8° occurred From the sixth through the eighth day the

temperature did not exceed 100° orally, and the patient remained afebrile for the remainder of her hospital stay For the first 2 days she received parenteral fluids and insulin By the third day she was placed on a 1200 Cal diet of 125 gm carbohydrate, 60 gm protein, and 40 gm fat, which was increased to 1800 Cal as 180 gm carbohydrate, 80 gm protein, and 84 gm fat when her appetite returned to normal A fasting blood sugar was 95 mg per cent on April 22 With diet and 10 U of NPH insulin each morning before breakfast she remained aglycosuric and was discharged as improved on this regimen on May 7, 1958

After her complete recovery, insulin was discontinued and tolbutamide therapy was begun on June 21, the patient receiving 3 0 gm the first day, 2 0 gm the second day, and 1 0 gm daily thereafter She remained asymptomatic Weekly blood counts, urinalyses, thymol turbidity tests, cephalin flocculation tests, A/G ratios alkaline phosphatase tests, and total and direct serum bilirubin values remained normal Tolbutamide was continued for 7 weeks until August 9, 1958, when chlorpropamide was substituted at a dosage of 250 mg daily On this reduced dosage of chlorpropamide she has remained asymptomatic and aglycosuric, with normal blood glucose levels for the 5 weeks she has been on the drug Weekly blood counts, urinalyses, thymol turbidity tests, cephalin flocculation tests, A G ratios, alkaline phosphatase tests and total and direct serum bilirubin values have been determined and have been found to remain within normal limits

Case 2 O D a 68 year old Negro female was discovered to have diabetes mellitus in December 1955, with the typical symptoms of polyuria, polydipsia, polyphagia a 15-lb weight loss lack of energy, and pruritus vulvae for 8 months Her diabetes was treated with diet and 30 U of NPH insulin each morning before breakfast On October 3 1956 she was admitted to the Surgical Service of Grady Memorial Hospital for cholecystectomy At the age of 26 years she had had typhoid fever followed by jaundice In April 1956 the patient developed right upper quadrant pain and associated nausea and vomiting Cholecystogram at this time revealed nonopacification of the gall bladder and cholelithiasis Because of continued symptoms of indigestion and intermittent attacks of upper abdominal discomfort with radiation into the right scapular region cholecystectomy was performed Although her sclerae were thought to be slightly icteric on October 4 1956 the total serum bilirubin was 0 3 mg per cent with a direct bilirubin of 0 2 mg per cent At the same time the total protein was 6 4 gm , albumin 3 1 gm and globulin 3 3 gm The A G ratio was 0 94 thymol turbidity was 4 U and the cephalin flocculation test was 3+ These tests were repeated on October 5, 1956 The total serum protein was 7 6 gm albumin, 3 1 gm and globulin, 4 5 gm The A G ratio was 0 69, thymol turbidity was 4 U and the cephalin flocculation test was 2+ The patient withstood cholecystectomy uneventfully and was discharged from the hospital October 18, 1956 on an 1800 Cal diet of 175 gm carbohydrate, 70 gm protein, and 91 gm fat with 35 U of NPH insulin each morning before breakfast Her case was followed at the diabetes clinic and, on January 4, 1957, the NPH

and chlorpropamide therapy was instituted with a dose of 0 5 gm twice daily The day before a complete blood count was normal urinalysis was negative (except for a 4+ sugar) liver function tests were normal the thymol turbidity test was 4 3 U the alkaline phosphatase test was 3 7 Bodansky units and the total serum bilirubin was 0 35 mg per cent with a direct bilirubin of 0 15 mg per cent

Initially the patient tolerated the medication satisfactorily and was managed with the same degree of control as with the previous insulin regimen On the fourteenth day of chlorpropamide therapy the patient developed nausea and vomiting and a dark urine that persisted for 3 days (until April 18 1958) when she was admitted to the hospital Careful questioning revealed no history of exposure to rats or hepatotoxic agents or to anyone with hepatitis Needle punctures were limited to her own insulin syringe and needle used by no one else and to those done in our laboratory with careful precautions

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icteric the lungs were clear and the heart sounds were normal Abdominal examination revealed a cholecystectomy scar The liver edge was felt two *finger breadths below the right costal margin and was nontender* The remainder of the examination was noncontributory The urinalysis on admission was negative except for the presence of bile the blood count was within normal limits Liver function tests on April 18 1958 revealed a total *protein of 7 3 gm albumin 3 4 gm globulin 3 9 gm* and an A/G ratio of 0 87 The thymol turbidity test was 4 2 U and the cephalin flocculation test was 1+ The alkaline phosphatase test was elevated to 8 5 Bodansky units The serum bilirubin was elevated to 6 35 mg per cent with a direct bilirubin *of 4 25 mg per cent*

The prothrombin time of the patient was 39 2 sec with a control value of 11 4 sec at 100 per cent 19 8 sec at 30 per cent and 26 0 sec at 20 per cent Following the intravenous administration of 50 mg of vitamin K the prothrombin *time on April 19* was 15 0 sec with a control value of 12 4 sec at 100 per cent 21 0 sec at 30 per cent and 27 6 sec at 20 per cent On April 21 it was 14 2 sec with the same control values as on April 19 On April 28 it was 13 4 sec with a control value of 13 4 sec at 100 per cent *21 0 sec at 30 per cent and 29 0 ec at 20 per cent* On April 22 1958 the

it was 2 8 Bodansky units Weekly determinations since ... normal

On April 22 1958 there was an increase in reversal of the A/G ratio, with a total protein of 7 2 gm albumin 3 1 gm globulin 4 1 gm and an A/G ratio of 0 76 On April 25 serum electrophoresis revealed a nonspecific increase in globulins On April 28 the total protein was 7 7 gm albumin 4 5 gm globulin 3 2 gm and an A/G ratio of 1 4 Weekly A G ratios have remained normal A Bromsulphalein test on April 28 revealed 10 per cent retention of dye after 45 min This was repeated on July 15 1958 with a retention of 4 per cent

A liver biopsy was obtained on April 23 1958 after the patient had almost completely recovered The biopsy revealed minimal hepatocellular degeneration minimal parenchymal cell jaundice and minimal pericholangitis with bile stasis

A chest X ray was negative While in the hospital the patient remained afebrile By the following day the nausea and vomiting had subsided and a sense of well being returned The diabetic state was well controlled on an 1800 Cal diet of 175 gm carbohydrate 70 gm protein 91 gm fat and 30 U of NPH insulin each morning before breakfast She was discharged without complaints on April 28 1958

One month after complete recovery insulin was discontinued tolbutamide therapy was begun on June 1 1958 in dosages of 3 0 gm the first day 2 0 gm the second day and 1 0 gm daily thereafter for 7 weeks Weekly blood counts urinalyses thymol turbidity tests cephalin flocculation tests A G ratios alkaline phosphatase tests and total and direct serum bilirubin values have remained normal The cephalin flocculation test was 3+ on 2 occasions but negative to 2+ in the remaining tests She remained asymptomatic and fairly well controlled on this regimen On July 19 1958 tolbutamide was discontinued and chlorpropamide was begun with a daily dosage of 250 mg For 8 weeks she has remained asymptomatic and is now essentially aglycosuric On August 28 1958 the fasting blood sugar was 137 5 mg per cent and a blood sugar 2 hours after breakfast was 175 mg per cent Weekly blood counts urinalyses cephalin flocculation tests and total and direct serum bilirubin values were normal

Case 3 L C a 50 year old Negro female was seen at the eye clinic in December 1955 complaining of difficulty with vision in the left eye She was found to have bilateral cataracts When she was admitted on January 9 1956 for cataract extraction glycosuria was found and the fasting blood sugar was 282 mg per cent Repeat fasting blood sugars were 313 mg per cent and 264 mg per cent On a 1500-calorie diet of 160 gm carbohydrate 55 gm protein and 70 gm fat she developed ketosis before insulin treatment was started After cataract extraction was accomplished on the left eye she was discharged [illegible]
of NPH insulir [illegible]
clinic At thi [illegible]
sugar Since [illegible]
min the insulin dosage was reduced to 15 U of NPH with the continuation of a 1500 Cal diet She maintained fairly good control of her diabetes on this regimen with occasional glycosuria On January 16 1957 cataract

extraction was performed on the right eye The patient was observed at occasional intervals and seemed to be under fairly good control with 15 U of NPH insulin each morning the weight varied usually between 100 and 105 lb On March 18, 1958 insulin was discontinued and the patient was

At this time the total serum bilirubin of 0 14 mg per cent, phosphatase was 4 87 Bodansky units On March 28 the patient was aglycosuric and was feeling well, her weight was 105 lb On April 18, 1958 anorexia, nausea, vomiting and generalized pruritus appeared and dark urine was noted for 2 or 3 days Chlorpropamide was discontinued and 15 U NPH insulin each morning was again started on April 21, 1958 On April 25 1958 she stated that the gastrointestinal symptoms disappeared as soon as she stopped taking chlorpropamide and the urine became normal in color about two days later, there was still some pruritus and her weight was 98 lb The sclerae were mildly icteric and there was an erythematous maculopapular rash on the trunk The liver and spleen were not definitely palpable Careful questioning revealed no history of exposure to rats or hepatotoxic agents or to anyone with hepatitis Needle punctures were limited to her own insulin syringe and needle, used by no one else, and to those done in our laboratory with careful precautions On April 28 the total serum bilirubin was 1 85 mg per cent, with a direct bilirubin of 0 9 mg per cent The thymol turbidity test was 1 45 U, alkaline phosphatase test was 13 2 Bodansky units, cephalin flocculation test was 1+, and A/G ratio was 0 92 The prothrombin time was normal All symptoms and signs cleared rapidly so that on April 30, 1958 she appeared well, except for slight scaling of the skin over the areas of the abdomen previously involved by the rash There was occasional glycosuria on 15 U of NPH insulin daily and the weight was 98 lb On May 30 the

Bodansky units A/G ratio was 1 41 1 cephalin flocculation was 1+ prothrombin time was normal and the BSP test showed 2 per cent retention

tests A/G ratios alkaline phosphatase tests and total and direct serum bilirubin values have remained normal

Discussion

Although we can not rule out absolutely the coexistence of a viral hepatitis as the cause of the jaundice the presence of an erythematous maculopapular rash in all 3 patients and the pre ence of a 13 per cent eosinophilia in 1 patient suggest very strongly that the etiology of the jaundice was related to a hypersensitivity reaction to chlorpropamide Many drugs have been stigmatized as producing jaundice Probably the drug that now most commonly causes jaundice is chlorpromazine J H Moyer *et al*[4] reported 1 case of obstructive jaundice without hepatocellular involvement in 500 patients treated L L Hollister[5] reported 17 cases of jaundice due to chlorpromazine among a total of more than 900 patients treated with the drug He estimates that the over all incidence of jaundice in patients treated for at least 1 week with chlorpromazine is probably between 1 and 2 per cent The work of these investigators strongly suggests a hypersensitivity reaction as the cause of the hepatic complications The clinical picture of fever gastrointestinal symptoms malaise and jaundice developing within 4 to 5 weeks of therapy together with the laboratory findings primarily of an obstructive type of jaundice is very similar to the findings in our cases

Methyltestosterone as a cause of jaundice has also been described[6] A B Gutman[7] has discussed the drug reactions characterized by cholestasis associated with intrahepatic biliary tract obstruction He reported the following drugs as causative agents arsphenamine dinitrophenol cinchophen aurothioglucose methyltestosterone norethandrolone (Nilevar) thiouracil para aminosalicylic acid sulfadiazine methimazole 8 (para aminobenzyl) caffeine He considered these reactions to be the re ult of hypersensitivity to the drugs involved F A Gibbs and F W Stamps[8] report serious liver damage occurring in about four persons in one thousand receiving phenurone

used as chemotherapeutic agents have been well known for some time[9,10] It is thought that a hypersensitivity reaction is responsible for these side effects although the exact mechanism of action on the liver i not definitely known

The predominant picture in our cases was an obstructive jaundice with hyperbilirubinemia an elevated alkaline pho phatase and only slight abnormalities in hepatocellular function It was interesting that two of the

patients had a marked elevation of the prothrombin time Another evidence that serious hepatocellular damage had not occurred was the prompt return to normal of the prothrombin time after vitamin K_1 therapy Cholestasis was a prominent feature of the liver biopsy findings although hepatitis was also present The similarity of our findings to the reports of hypersensitivity reactions with jaundice due to various drugs makes us feel that our cases definitely have this basis

Because of a rather high incidence of cross sensitivity to the sulfonamides we attempted to determine whether these patients would show any side effects when placed on tolbutamide therapy With an initial dose of 3 0 gm a second dose of 2 0 gm and a daily dose of 1 0 gm there was no evidence of hypersensitivity The patients remained asymptomatic, and weekly blood counts urinalyses and liver function tests remained normal for the 7 weeks they were kept on tolbutamide

After tolbutamide therapy these patients were again placed on chlorpropamide but with a reduction of the dose from 1 0 gm to 250 mg daily Two of the 3 have tolerated the drug without side effects for 5 and 8 weeks respectively They have remained asymptomatic Weekly blood counts urinalyses and liver function tests have remained normal On the ninth day of resumption of chlorpropamide the third patient noticed nausea and vomited once She immediately discontinued the drug on her own volition and resumed tolbutamide which she has continued to take without incident At the time she stopped chlorpropamide no abnormalities by physical examination blood count urinalysis or liver function tests could be found We are not in a position to state whether these gastrointestinal complaints were related to the drug or were due to some unrelated cause One might expect evidence of hypersensitivity when the patients were again administered the drug In 1 of these patients it may have been the cause of the gastrointestinal complaints Possibly the reduction in dosage may have decreased the likelihood of this reaction With true hypersensitivity however one would have expected some evidence of hypersensitivity even with a smaller dose of the drug L E Hollister[5] reported recurrence of jaundice in 6 of 11 patients treated with chlorpromazine 9 of the 11 revealing some evidence of hypersensitivity None of his patients showed cross sensitivity to chlorpromazine

Summary

Three cases of jaundice as a hypersensitivity reaction to chlorpropamide have been presented

The jaundice was primarily an obstructive type with hyperbilirubinemia and an elevated alkaline phosphatase

There was no evidence of cross sensitivity to tolbutamide

No recurrence of jaundice or evidence of impairment of hepatic function occurred when a smaller dose of chlorpropamide was given

On the ninth day of resumption of chlorpropamide one case developed mild gastrointestinal symptoms These symptoms cannot be definitely related to the drug

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STUDIES ON THE USE OF CHLORPROPAMIDE IN PATIENTS WITH DIABETES MELLITUS*

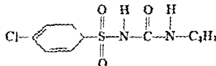
N Sheldon Skinner Jr ,† Robert L Hayes,‡
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The study of oral hypoglycemic agents has greatly stimulated research in diabetes mellitus Many new compounds capable of producing hypoglycemia are being synthesized and studied in an effort to find one such compound that will prove to be as effective as insulin in controlling diabetes mellitus and yet still be suited to oral administration

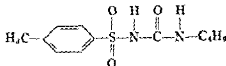
It is the purpose of this report to describe our clinical experience with chlorpropamide, a halogenated sulfonylurea compound, and to compare in detail the efficiency of this drug with that of insulin in managing patients with diabetes mellitus

Chemistry and Pharmacology

Chlorpropamide has the chemical name of 1 propyl 3 (*p*-chlorobenzene sulfonyl)urea and the chemical structure



Tolbutamide, in comparison has the chemical name of 1-butyl 3 tolyl-sulfonylurea and the chemical structure



The LD_{50} in mice and rats is comparable with both chlorpropamide and tolbutamide according to one study, while another group reports that chlorpropamide shows greater toxicity Both of these compounds showed similar side effects (depression, ataxia, tremors) in mice and rats, usually only at the higher dose levels [1,2]

Acute, subacute, and chronic toxicity studies of this drug done on mongrel

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dogs showed no significant anatomical changes either gross or histological, according to one group of investigators whereas a second group found liver degeneration and fatty infiltration as well as nephrotic changes in the kidney [1, 2]

Blood level determinations were done on normal human volunteers Three groups of these volunteers were used equal numbers of volunteers were placed on dose levels of 0 25 gm per day 0 50 gm per day and 1 0 gm per day of chlorpropamide Each of these groups was observed to reach a mean plateau blood level of chlorpropamide on approximately the fifth day of drug ingestion The volunteers receiving 0 25 gm per day reached

10 mg per cent those receiving
cent and tho e receiving 1 0 gm
it [1]

per day of chlorpropamide became hypoglycemic thereby necessitating discontinuance of the compound Blood levels were determined in these three after drug withdrawal and it was found that the removal of chlorpropamide from the blood stream was accomplished very slowly suggesting that no marked active destruction of this compound occurs in the human body and that lo s from the blood stream is accompli hed by very slow renal excretion

Materials and Methods

Fifty two patients ranging in age from 25 to 88 years and with proved diabetes mellitus were maintained on chlorpropamide for periods ranging from 1 to 25 weeks Forty eight patients were ob erved at weekly or bimonthly intervals depending on the need for a visit

Four patients were carefully studied in the hospital on calculated diets Four others had been on no previous treatment prior to the administration of chlorpropamide Forty patients had been previously managed with insulin therapy five with tolbutamide and four by diet alone Only two patients had been treated for diabetic acidosis prior to administration of chlorpropamide

The selection of patients for the study was random although in some instances patients were begun on chlorpropamide because of frequent insulin reactions poor control with insulin inability to self administer insulin or insufficient funds with which to purchase insulin (FIGURE 1)

All patients were placed on calculated diabetic diets but only those who were evaluated during ho pital admi sion had careful control of diet The clinic patients who constituted the lower economic stratum of the patients evaluated did not observe rigid control of their diets

Clinic patients were evaluated for degree of control with chlorpropamide

urine examinations All blood sugars were determined by the Folin Wu method [3]

Twenty three patients had serum protein bound iodine (PBI) determina

tions made before or within seven days following the administration of the initial dose of chlorpropamide repeat PBI determinations were continued throughout the treatment period The PBI determinations were made by a modification of the alkaline ash method of Barker [4]

Twenty two patients had serum cholesterol determinations made before or within seven days following the administration of the initial dose of

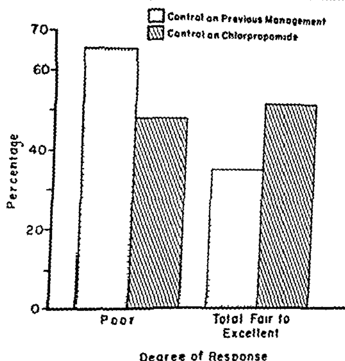


FIGURE 1 Comparison of the control of diabetes by chlorpropamide with previous management

chlorpropamide These determinations were made using the method of Pearson [5]

All patients had weekly to bimonthly urinalyses testing for red and white cells albumin specific gravity sugar and acetone Three patients had repeated liver function tests using standard methods that is alkaline phosphatase cephalin flocculation and thymol turbidity Four patients were evaluated with BSP tests All patients were observed with total and differential white blood cell counts as well as packed cell volumes or hemoglobin determinations

Initially all patients were begun on 2 gm of chlorpropamide daily The dose level was then quickly decreased to 1 0 or 0 5 gm per day The maintenance dose in the majority of patients treated was 0 5 or 0 25 gm per day and never exceeded 1 0 gm per day for any patient The various degrees of control of the diabetes mellitus in each patient both before and after chlor

propamide was administered were designated as follows: excellent, fasting blood sugar of 120 mg per cent or below; good, 120 to 140 mg per cent; fair, 140 to 160 mg per cent; and poor, more than 160 mg per cent.

Results

Effect on diabetic control. The effectiveness of chlorpropamide therapy in controlling diabetes mellitus was evaluated on the basis of the reduction in blood sugar level. The degree of blood sugar control was evaluated according to the age of onset of the diabetes, the amount of insulin required prior to

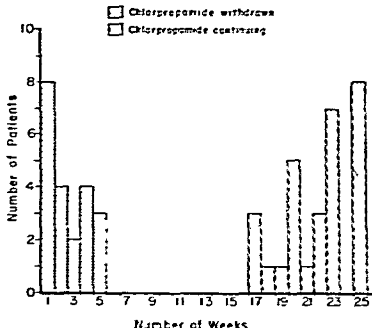


Figure 2. Duration of therapy with chlorpropamide.

treatment with chlorpropamide, the duration of diabetes prior to chlorpropamide treatment, and the age at which chlorpropamide treatment was begun.

Figure 2 shows the duration of chlorpropamide treatment in all patients studied. Of [illegible] patients begun on chlorpropamide, 22 ([illegible] per cent) were stopped by the end of [illegible] weeks of treatment, and [illegible] ([illegible] per cent) were maintained on treatment for 17 to 25 weeks. It should be noted that no patient was forced to withdraw from chlorpropamide therapy after 5 weeks of treatment. Of the 22 patients forced to stop chlorpropamide treatment, 6 had poor control of blood sugar without significant side reactions, 13 had poor control of blood sugar with significant side reactions, and 3 had fair-to-excellent control with significant side reactions.

Figure 1 shows graphically the poor and the fair-to-excellent control

attained on the various means of prechlorpropamide treatment and on chlorpropamide treatment

TABLE 1 shows the degree of control of diabetes attained with chlorpropamide after 25 weeks of treatment. The over all control was fair to-excellent in 54 per cent and poor in 46 per cent of patients. While receiving various

TABLE 1
DEGREE OF CONTROL OF DIABETES OBTAINED WITH CHLORPROPAMIDE AFTER 25 WEEKS OF TREATMENT

Age groups	Control				
	Excellent	Good	Fair	Poor	Total
21 to 30				1	1
31 to 40				2	2
41 to 50	2	1	1	3	7
51 to 60	6	2	1	4	13
61 to 0	7	1	4	8	20
71 to 80	1	2		5	8
81 to 90				1	1
Total	16	6	6	24	52
Per cent total	31 0	11 5	11 5	46	

TABLE 2
CORRELATION OF BLOOD SUGAR CONTROL WITH THE AGE OF ONSET OF DIABETES MELLITUS

Age group (years)	Number of patients	Number of patients showing fair to excellent blood sugar response	Percentage of response
11 to 20	1	0	0
21 to 30	3	0	0
31 to 40	4	1	25
41 to 50	11	8	73
51 to 60	15	9	60
61 to 70	13	9	69
71 to 80	5	1	20

means of treatment prior to chlorpropamide administration, only 34 per cent of the 52 patients showed a fair to excellent response

TABLE 2 shows a correlation of blood sugar control with the age of onset of diabetes mellitus. The 51 to 60 and 61 to 70 age groups had approximately the same percentage of fair to-excellent responses, with the 41 to 50 age group showing the best response

TABLE 3 shows a correlation of blood sugar response with the duration of

iabetes prior to chlorpropamide treatment There is essentially an equal umber of patients in each group up to the 15 year duration group Because f the small number in the groups after this, no accurate evaluation can be nade The 0 to 1 year and 1- to 5 year duration groups showed, in effect, n equal degree of control, as did the 6- to 10 and 11- to 15-year duration roups

TABLE 3
CORRELATION OF BLOOD SUGAR RESPONSE WITH THE DURATION OF DIABETES PRIOR TO CHLORPROPAMIDE TREATMENT

Duration of diabetes (years)	Number of patients	Number of patients showing fair to excellent blood sugar response	Percentage of patients responding
Less than 1	16	9	54
1 to 5	14	10	72
6 to 10	9	2	22
11 to 15	10	5	50
15 to 20	1	1	100
20 to 25	1	1	100
30 to 35	1	0	0

TABLE 4
CORRELATION OF BLOOD SUGAR RESPONSE WITH AGES AT WHICH CHLORPROPAMIDE TREATMENT WAS BEGUN

Age group (years)	Number of patients	Number of patients showing fair to excellent blood sugar response	Percentage of response
21 to 30	1	0	0
31 to 40	2	0	0
41 to 50	7	4	57
51 to 60	13	9	70
61 to 70	20	12	60
71 to 80	8	3	38
81 to 90	1	0	0

TABLE 4 shows a correlation of blood sugar response with the ages at which chlorpropamide treatment was begun Four of the age groups shown have approximately the same number of patients, and all have essentially the same blood sugar response, with a fair to excellent response ranging from 38 to 70 per cent

TABLE 5 shows a comparison of blood sugar control while on chlorpropamide with that obtained on previous therapy Again, there is a general tendency that suggests that the lower the previously required insulin dose, the better was the response to chlorpropamide A 100 per cent response to

chlorpropamide was noted in the five patients previously controlled on tolbutamide

One hospitalized patient was carefully studied with both chlorpropamide and tolbutamide administration During the testing of chlorpropamide the patient was observed with PBI determinations, cholesterol determinations, fasting, 11 A M, 2 P M, and 4 P M blood sugars, radioactive iodine[131] (RAI) uptakes, and liver function tests This patient was an obese 65 year old Negro male with a history of diabetes mellitus of 20 years' duration He had been well controlled on 35 U of NPH U-80 insulin per day when he was first discovered to have diabetes mellitus He was then maintained on doses

TABLE 5
COMPARISON OF BLOOD SUGAR CONTROL WHILE ON CHLORPROPAMIDE WITH THAT OBTAINED ON PREVIOUS THERAPY

Previous means of treatment	Number of patients	Number of patients showing fair to excellent blood sugar response	Percentage of patients responding
No insulin	9	4	45
1 to 20 U insulin	9	6	67
21 to 50 U insulin	13	7	54
51 to 100 U insulin	13	5	40
101 to 200 U insulin	2	1	50
Over 200 U insulin	1	0	0
Tolbutamide	5	5	100

of tolbutamide ranging from 1 to 4 gm per day, although excellent control required 4 gm daily

FIGURE 3 shows the comparison of the control of this patient on tolbutamide and chlorpropamide after 10 weeks of study during each period During both these 10 week periods dietary control was essentially the same FIGURE 3 also shows that tolbutamide in doses of 4 gm per day was quite effective in maintaining the patient's blood sugar control in the range of excellent (below 120 mg per cent) while only a fair degree of control could be attained on 3 0 gm of tolbutamide per day No untoward reactions to tolbutamide were recorded during this time Also shown is the fact that

effective levels of the drug Three weeks after the drug was discontinued the fasting blood sugar levels rose from 100 to 150 mg per cent and by the fifth week had risen to 163 mg per cent It was concluded that the duration of action of chlorpropamide was approximately 3 weeks, and that normal blood sugar levels could again be attained within 1 week after starting 0 25 gm of chlorpropamide per day

The first dose of 3 0 gm of chlorpropamide reduced this patient's blood sugar level from 110 mg per cent to 63 mg per cent in 5 hours On the third day of therapy at this dose level a rather marked hypoglycemic reac

tion was produced during the early morning hours, with a blood sugar level of 57 mg per cent The diabetes was well controlled on maintenance levels of 0 25 gm per day

Mild side reactions were produced by 3 0 gm of chlorpropamide per day They included minimal confusion and weakness, moderate somnolence, and minimal anorexia that persisted for 2 to 3 days following reduction of chlorpropamide to 0 25 gm per day but rapidly disappeared thereafter There

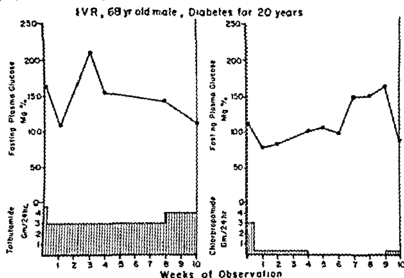


FIGURE 3 Comparison of the effect on fasting blood sugar levels of tolbutamide and chlorpropamide

were no side reactions of any kind produced by the maintenance dose of 0 25 gm daily

The Effect of Chlorpropamide on Serum Cholesterol Levels

Serum cholesterol levels were determined on twenty two patients receiving therapeutic doses of chlorpropamide All patients were placed on calculated diabetic diets TABLE 6 shows the serum cholesterol levels obtained at various times from this group

It is of interest that the serum cholesterol level of 9 (41 per cent) of the 22 patients decreased by 50 mg per cent or more while under treatment with chlorpropamide The mean initial serum cholesterol level was 280 mg per cent and the mean level after 25 weeks of treatment was 239 mg per cent—a mean decrease of 41 mg per cent, which includes the mean decreases in 13 patients who did not show a decrease of 50 mg per cent or more There was a mean decrease of 79 mg per cent in the 9 patients who showed a 50 mg per cent or more decrease in serum cholesterol levels The difference between these means was not statistically significant

In almost all cases, the higher the serum cholesterol level prior to chlor

propamide administration the greater the fall in serum cholesterol level following chlorpropamide therapy Evidence of this effect was further substantiated by the observation that in the 9 patients who showed a decrease in serum cholesterol of 50 mg per cent or more the mean serum cholesterol level prior to chlorpropamide therapy was 324 mg per cent in the 13 patients showing a decrease in serum cholesterol level of less than 50 mg per cent the mean initial level was 250 mg per cent There was no correlation

TABLE 6
SERUM CHOLESTEROL DETERMINATIONS MADE AT VARIOUS WEEKS AFTER STARTING CHLORPROPAMIDE THERAPY

Patient	Pretreatment level 1 wk	Level weeks after therapy 2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	21	Mean following therapy
1	222		231	205		253			231				248								234
2	304	272		226	225																241
3	352											296									296
4		290	255							192					218			242		221	225
5	324							303		243						309					265
6	238					200		181					206		194		179			177	189
7	306				224								147								185
8		294								256										244	250
9	252		245									237		249							243
10	266						283		257	276	275			276				275			273
11	266		257		214			254				279			246		213			184	235
12	261										239									235	237
13	207		172	229		190						250			245						217
14	270			224		228		179			271			291							238
15	220					268		213				241		222						235	235
16	248								254		247										250
17	318			250			288	273		243								209			252
18	194			193				157	129				170								162
19	290					223	212		213			246							190		216
20	274	264		222						212			210							261	234
21	471			339				349	347												345
22	288				181	269															225
Mean	280																				239

between the degree of blood sugar control and the fall in serum cholesterol level nor was there a correlation between the fall in serum cholesterol the duration of diabetes or the amount of insulin previously required to control the diabetes

Effect of Chlorpropamide on Thyroid Function

Twenty three patients had repeated serum PBI levels while on chlorpropamide therapy TABLE 7 shows the PBI determinations made at various weeks after starting chlorpropamide The initial mean PBI level prior to chlorpropamide therapy was 5 8 μg per cent, and the mean level following chlorpropamide therapy was 5 4 μg per cent—a mean decrease of 0 4 μg per cent The difference in these means was not statistically
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TABLE 7
SERUM PROTEIN BOUND IODINE DETERMINATIONS MADE AT VARIOUS WEEKS AFTER STARTING CHLORPROPAMIDE THERAPY

Patient	Pre treatment level 1 wk	Level weeks after therapy																					Mean following therapy
		2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	18	19	20	21	22	23	
1	5 5						4 4	5 8		4 6				3 1	5 6	3 5							4 5
2	7 5	5 4						5 3		7 6					4 4	6 0				4 6			5 6
3	5 1	5 1										5 8		5 4									5 4
4		5 2							5 7			6 5											6 1
5	6 3			6 3																			6 3
6	6 3			5 8						6 7		5 6											6 0
7	6 3											6 5		4 4					5 1				5 3
8		5 3	6 8	7 4		3 6						5 2											5 8
9	6 7	6 3		4 9				7 7				6 6						4 1					5 9
10	7 2								6 6														6 6
11	6 0										6 0		5 1					4 3					5 1
12	5 6		4 4								4 9			2 4								2 8	3 6
13	6 7							3 7		6 1								3 7					4 5
14		6 4							7 9		6 4								7 5				7 3
15	4 5						5 7		7 5		5 4	5 3								4 4			5 7
16		6 5		5 8																			5 8
17	4 5				4 6				5 2														4 9
18		5 5	3 5		5 8	3 9	4 2				4 7			5 2									4 4
19	5 9	5 9	5 2			5 0			5 8				6 2										5 6
20		3 3						4 4				5 8					7 3						5 8
21	6 3			6 3																			6 3
22						5 7		5 4									3 3						4 4
23	5 9	3 2	2 5				4 7			5 2	5 8	5 2									3 9		4 4
Mean	5 8																						5 4

Several cases warrant discussion because of marked decreases in their serum PBI levels while on chlorpropamide therapy One patient showed a gradual decrease in serum PBI levels from 5 6 μg per cent to 3 6 μg per cent, with levels falling to less than 3 μg per cent on 2 occasions A second patient showed a gradual decrease in serum PBI from an initial level of 6 0 μg per cent to 4 3 μg per cent after 19 weeks of therapy Seven other patients showed a decrease in PBI levels of 1 0 μg per cent or more while on chlorpropamide treatment As far as could be determined, no symptoms referable to these decreases in PBI levels were produced

In an effort to determine whether chlorpropamide depressed the 24 hour uptake of RAI, 5 patients had control 24 hour uptakes and uptakes following administration of large doses (3 to 7 gm) of chlorpropamide RAI uptakes were also determined at hourly intervals and, when a plateau was reached, potassium thiocyanate (1 0 to 1 5 gm) was administered The mean prechlorpropamide 24 hour RAI uptake in the 5 patients was 18 1 per cent, and the mean 24 hour uptake following chlorpropamide administration was 11 4 per cent—a mean decrease of 6 7 per cent The difference between means was not statistically significant There was no significant discharge of RAI following the administration of potassium thiocyanate in any of the patients It appears, then, that chlorpropamide, at least in large doses, may be capable of suppressing the 24 hour RAI uptake but there is no evidence to suggest that chlorpropamide exerts any interfering action on the peroxidase enzyme system in a manner similar to propylthiouracil

Effect of Chlorpropamide on Blood and Urine

All patients had complete peripheral blood evaluations and complete urinalyses at weekly intervals when first started on chlorpropamide therapy, the frequency of these determinations was decreased as therapy was prolonged

At no time was there any evidence of anemia or granulocytopenia, although in a number of patients a slight increase in lymphocytes and monocytes was noted at approximately the same time the blood sugar fell toward normal No pathological urine findings that could be attributed to chlorpropamide were noted in any of the patients

Effect of Chlorpropamide on Liver Function

Three patients who had been maintained on relatively large doses of chlorpropamide for prolonged periods were evaluated with cephalin flocculation, thymol turbidity, and alkaline phosphatase tests Four patients who had also been maintained for prolonged periods on large doses of chlorpropamide were evaluated with BSP tests In none of these patients could any evidence of liver damage be found Jaundice was not a finding in any of the fifty two patients treated

Untoward Effects

Although unpleasant to incapacitating side reactions occurred on all dose levels of chlorpropamide employed (0 25 gm every other day to 3 0 gm per

day), reactions were sufficiently severe in only 2 patients on 0 25 gm per day to warrant discontinuance of medication

TABLE 8 shows the percentage of side reactions in patients receiving various amounts of medication There was an over all incidence of side effects of some degree in 37 per cent of the patients, but only 15 per cent of the patients had sufficiently severe reactions to necessitate withdrawal of medication

TABLE 9 reveals that weakness, hunger, dizziness, and somnolence were the most common complaints occurring on all dose levels used The severity

TABLE 8
PERCENTAGE OF SIDE REACTIONS IN PATIENTS RECEIVING VARIOUS AMOUNTS OF CHLORPROPAMIDE

Dose level* per day (gm)	Number of patients	Number of patients having side reactions†	Percentage
3 00	3	2	67
2 00	13	12	92
1 50	2	1	50
1 00	13	7	54
0 50	39	24	62
0 25	32	15	47

* Several patients are included at more than one dose level
† The side reactions in patients receiving 0 50 gm or less of chlorpropamide were usually mild and relieved by food

TABLE 9
ORDER OF FREQUENCY OF SIDE REACTIONS OBSERVED WITH CHLORPROPAMIDE THERAPY

1 Dizziness
2 Weakness
3 Hunger
4 Somnolence
5 Ataxia
6 Nausea
7 Skin eruption
8 Confusion
9 Headache
10 Anorexia
11 Vomiting
12 Acidosis
13 Hypoglycemic coma

of each of these 4 symptoms was related directly to the size of the dose used the larger the dose, the more severe the complaint These symptoms could not be correlated with hypoglycemia in all instances but frequently—particularly in patients receiving 0 25 to 0 50 gm of chlorpropamide per day symptoms could be correlated with low blood sugar levels Symptoms clinically resembling hypoglycemia could not be relieved by food ingestion when they were produced by 2 0 gm per day or more of chlorpropamide, but could be relieved effectively by food ingestion when produced by doses of from 0 25 to 0 50 gm per day Dizziness, weakness, ataxia, somnolence and confusion usually occurred together, and at no time were they the sole complaints In all cases these side reactions were most severe when they occurred on dose levels of from 1 0 to 3 0 gm of drug per day, when produced

by doses of 0.5 to 0.25 gm. daily they were rarely incapacitating. These effects were usually produced within hours of drug ingestion when they accompanied doses of 1.0 *to 3.0 gm. daily*.

Severe somnolence was a complaint only in that group receiving 2.0 gm. or more of chlorpropamide daily. These patients in the absence of preoccupation would immediately fall asleep, although they could be quite easily aroused and were not confused following arousal. Weakness was a prominent complaint in 2 patients receiving doses of chlorpropamide of 0.50 gm. per day or less and could not be relieved by the ingestion of food.

Ataxia when it occurred, was usually present the day therapy was initiated but was maximal the second to third day of treatment. Many patients frequently fell as a result of it. Relief was always rapid and complete after marked reduction of the dose of chlorpropamide. Only 1 of 32 patients receiving 0.25 gm. daily complained of ataxia, in this patient, the condition persisted for 1 week following withdrawal of the drug.

Nausea and vomiting occurred primarily in patients receiving 1.0 gm. or more daily of chlorpropamide and was rapidly and effectively relieved by decreasing the dose used. At no time was vomiting sufficiently severe to [illegible] When nausea and vomiting [illegible] *mide daily*, *it was*

[illegible] ctions were commonly encountered in patients taking *0.25 to 0.50 gm. of chlorpropamide* daily. Of these reactions, headache and hunger were quite common and, even though they frequently occurred together they sometimes occurred independently. Characteristically they began late in the afternoon, just before the evening meal and would be completely relieved by the ingestion of food. Infrequently patients complained of being awakened by hunger during the early morning hours food ingestion immediately prior to retiring prevented this effect. Somnolence when it occurred in patients receiving 0.25 to 0.50 gm. of drug daily, frequently occurred after 6 to 8 *weeks of* therapy on chlorpropamide and appeared late in the afternoon, was associated with hunger and dizziness and was readily relieved by food.

Skin rash and pruritis were very common reactions, occurring in 12 of 71 patients (17 per cent) receiving 0.25 to 0.50 gm. daily of chlorpropamide [illegible] erythema multiforme while being treated. A definite [illegible] ide therapy and the [illegible] otion was seen infre[illegible] ly, as only 2 patients were maintained on such doses for more than 2 weeks. *These 2 patients* did not experience side reactions of any type, which suggests the marked individual variation of susceptibility to this drug.

Five of 39 patients receiving 0.5 gm. of chlorpropamide daily complained of skin rash. The length of time patients were maintained on such a *dose* was considerably longer than the length of time patients were maintained *on higher doses*.

Seven of 32 patients taking 0.25 gm. daily of chlorpropamide had skin rash

and pruritis at one time during the course of therapy Both of these effects were usually noticed within 3 weeks after initiation of therapy and the incidence of these complaints increased as the duration of chlorpropamide therapy increased

Skin rash characteristically was preceded or accompanied by pruritis although this was not a constant finding A small percentage of the patients complained of skin rash alone When pruritis preceded the skin rash it invariably did so by 1 to 3 weeks and did not persist following disappearance of the rash

The rash was usually a maculopapular eruption erythematous discrete but never confluent Characteristically the face (particularly the malar eminences) the neck the upper third of the chest (both anteriorly and posteriorly) and the proximal third of the arms were the areas included When pruritus developed first it was localized to these same areas One patient developed a localized pretibial eruption The skin rash was usually transient and of only 2 to 14 days duration disappearing completely after this time Two patients were exceptions to this one after being treated with 0 5 gm of chlorpropamide daily for 3 weeks developing an itching red rash Chlorpropamide therapy was immediately withdrawn but the rash continued to increase in severity and 4 weeks later a severe exfoliative dermatitis developed with profuse vaginal and urethral discharge labial ulcers and mouth ulcers The patient became extremely ill although following hospitalization for 2 weeks and extensive treatment she did well and was discharged without sequelae A second patient was treated with 0 5 gm of chlorpropamide daily for 1 week The drug was then withdrawn because of the development of nausea weakness ataxia and dizziness One week after discontinuance of the drug the patient developed an itching, urticarial type of eruption that responded only minimally to extensive antihistaminic treatment The rash progres ed for 5 weeks and disappeared by the end of 7 weeks recovery again was complete

Acidosis occurred in only two patients neither of whom had a blood sugar response to chlorpropamide In both patients it developed two days after treatment was started One patient was a juvenile diabetic with a past history of acidosis the other an adult diabetic without such previous history

Hypoglycemic coma occurred in 1 patient a 60 year-old Negro male who had previously been poorly controlled on 130 U of NPH U 80 insulin daily At the time of development of hypoglycemic coma he was receiving 1 0 gm of chlorpropamide per day Symptoms developed during the early morning hours after he had failed to eat the evening meal the preceding night On admis ion to the emergency room his blood sugar was 30 mg per cent He re ponded immediately to intravenous glucose and has been well controlled since on 0 5 gm of chlorpropamide per day without further complaint

Death occurred in one patient being treated with chlorpropamide but as this patient had her terminal treatment in an outlying hospital the details surrounding her death are unknown An autopsy was not obtained but she had numerous severe complications such as congestive heart failure and chlorpropamide was thought not to be related to her death

Summary and Conclusions

Studies are reported on 52 randomly selected adult patients with diabetes mellitus who were treated from 1 to 25 weeks with 0 25 to 3 0 gm daily of chlorpropamide Thirty patients were maintained on this drug for from 17 to 25 weeks

Fair to excellent blood sugar control was obtained in 28 (54 per cent) of the patients treated The best response was obtained in those patients with a requirement of less than 21 U of insulin daily for control of the diabetes

One patient was treated with both chlorpropamide and tolbutamide and ... times as potent ... No untoward

A mean decrease in serum cholesterol level of 41 mg per cent was observed in twenty two patients following treatment with chlorpropamide This decrease was not statistically significant A tendency for serum cholesterol to be lowered by chlorpropamide when initial serum cholesterol levels were high was suggested The duration of diabetes previous control of diabetes and the amount of insulin previously required for control played little or no part in determining the response of cholesterol to chlorpropamide

Twenty three patients were studied to observe the effects of chlorpropamide on thyroid function A mean decrease in serum PBI levels of 0 4 μg per cent was observed following 25 weeks of treatment with chlorpropamide this was not statistically significant Five patients were observed to have a mean decrease of 6 7 per cent in 24 hour RAI uptakes following chlorpropamide treatment but again this was not statistically significant

Untoward effects were common with chlorpropamide treatment and in general tended to be most severe and most frequent on dose levels of 1 0 gm or more daily While these reactions were due to hypoglycemia in some instances and could be relieved by food ingestion in many instances they appeared to be the result of a direct effect of chlorpropamide Side reactions were also commonly observed on maintenance dose levels of 0 25 to 0 50 gm daily but these were usually relieved by the ingestion of food and were thus transient and of little consequence

Renal and hematological damage was not found with chlorpropamide therapy nor was liver function impairment observed

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CLINICAL EXPERIENCE WITH CHLORPROPAMIDE IN THE MANAGEMENT OF DIABETES MELLITUS*

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Introduction

This report is based upon our observations of the compound 1-propyl-3-(*p*-chlorobenzenesulfonyl)urea (chlorpropamide) orally administered as an agent for the control of glycosuria and hyperglycemia as tested in 49 patients with diabetes mellitus for periods of 2 to 5 months. The drug has been found to be effective for this purpose in 76 per cent of this group, most of whom were patients whose diabetes began in their fifth decade or later. The data have been analyzed in terms of an arbitrary system for grading the degree of control achieved with the aid of the drug, and an appraisal on this basis of clinical factors influencing the effectiveness of the drug has been made. The concentration of chlorpropamide in blood serum attained at various dosage levels has been determined also, and the relation of these two factors to the effectiveness and toxicity of the drug has been evaluated.

Clinical and Experimental Data

Of the 49 patients included in this study, 35 were females and 14 males. All but 6 of the patients previously had required insulin, and 14 of these had been treated with tolbutamide and had been adequately controlled by that compound. The remaining 6 patients were new diabetics, 2 having been controlled on tolbutamide prior to chlorpropamide administration; in all 6, the blood sugar levels had been constantly higher than 195 mg./100 ml. before drug treatment, and good control was obtained by treatment in all 6.

Ten of the patients were studied initially for several weeks as inpatients in the Special Study Unit of Jackson Memorial Hospital. During the study they were kept on constant diet, and the excretion of glucose and ketones in the urine was determined quantitatively on partitioned 24-hour urine collections. Fasting blood sugar was also determined daily. After a suitable control period of observation, chlorpropamide therapy was begun, usually as 0.5 gm. twice daily for two days, with subsequent maintenance on a single daily dose. Insulin, when it was being given during the control period, was withdrawn within 2 to 4 days of initiation of chlorpropamide therapy with initial daily decrements of about 50 per cent.

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were controlled only insofar as they adhered to their diet prescription. They were seen at weekly intervals until their drug therapy and the response thereto were stabilized, and subsequently at intervals of two to three weeks.

If good control was achieved with 500 mg. of chlorpropamide daily, the maintenance dose was lowered; if satisfactory control was not achieved with 500 mg. daily, the dose was increased.

Determinations of chlorpropamide concentration in blood sera were made in both outpatients and inpatients with the aid of the method of Toolan and Wagner.[1]

TABLE 1 summarizes the status of the 49 patients with respect to previous treatment.

TABLE 1
NUMBER OF PATIENTS ACCORDING TO PREVIOUS THERAPY

Insulin	29
Insulin then tolbutamide	14
Tolbutamide	2
New diabetics	4
Total	49

Criteria of Control

Criteria for satisfactory control were arbitrarily established as follows: good: fasting true blood sugar consistently below 120 mg./100 ml., with no glycosuria; fair: fasting blood sugar between 120 and 170 mg./100 ml., with glycosuria less than 0.5 per cent.

Analysis of the Data

Age at onset of diabetes. The effectiveness of chlorpropamide in controlling hyperglycemia is most striking in those patients in whom diabetes mellitus first made its appearance after the age of 40. Of 43 patients in this category who were given the drug, 36, or 84 per cent, were adequately controlled without insulin. The excellent response often observed in these patients is exemplified in FIGURES 1 and 2.

In 5 patients whose diabetes began between the ages of 20 and 39, however, therapy was successful in only 1. The total lack of response encountered in patients of this group is illustrated in FIGURE 3. Chlorpropamide was also ineffective in the 1 child among our patients who received the drug, a 14-year-old boy with diabetes of 5 years' duration who required 52 U. of lente insulin daily. TABLE 2 gives the age-at-onset data for the 49 patients. [illegible] of the

variation in the sample, for the number of patients is small and, if [illegible] is confined to 4 of these patients who had had diabetes more than 20 years, the drug is shown effective for 75 per cent.

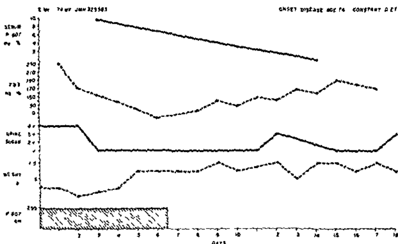


FIGURE 1 Response of a previously uncontrolled diabetic to chlorpropamide After a control period on a constant diet chlorpropamide therapy was begun without other

fasting blood sugar and P 607 to chlorpropamide

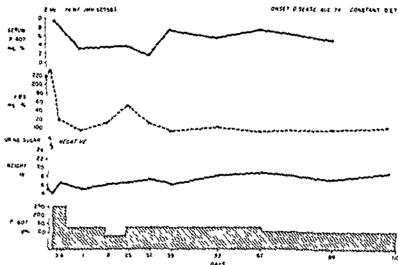
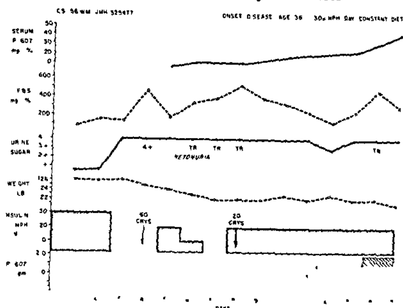


FIGURE 2 Effects of resumption of chlorpropamide therapy in the patient of FIGURE 1 Control for a 4½ month period has been excellent at a low dose level in this patient with newly diagnosed diabetes mellitus.



propamide (to 2 0 gm daily) supplemented by 20 U ...

Table 2
Relation of Age at Onset of Diabetes and Response to Therapy

	Age at onset, years				Total patients	Percentage
	0 19	20 39	40-59	60-80		
No of patients	1	5	22	21	49	—
Control						
Good	0	1	10	9	20	41 } 76
Fair	0	0	8	9	17	35 }
Unsatisfactory	1	4	4	3	12	24
Percentage satisfactory	0	20	82	86	—	—

Previous insulin requirement There was no doubt that the effectiveness of chlorpropamide was related to the previous insulin requirement (TABLE 4) Good control was obtained in all patients who had not previously required ... of patients

controlled Our experience with those requiring more than 40 U of insulin daily was limited to two patients One was the juvenile mentioned above,

the other a 68-year-old white female with an onset age of fifty, requiring 80 U of NPH insulin daily. On a daily dose of 500 mg. chlorpropamide per day, her insulin requirement was reducible to 50 U. of NPH daily, but no lower.

Comparison of tolbutamide and chlorpropamide. In 15 patients previously maintained on tolbutamide in doses of 0.5 to 2.5 gm., chlorpropamide was substituted (TABLE 5). Thirteen of these had been on insulin prior to the tolbutamide; 2 were new diabetics. The effective dose of chlorpropamide

TABLE 3
RELATION OF DURATION OF DIABETES AND RESPONSE TO THERAPY

	Duration of diabetes, years					Total patients	Percentage
	1	1–5	6–9	10–15	15		
No. of patients	7	18	6	12	6	49	—
Control							
Good	5	8	2	2	3	20	41 }76
Fair	0	6	4	7	0	17	35
Unsatisfactory	2	4	0	3	3	12	24
Percentage satisfactory	71	78	100	75	50	—	—

TABLE 4
RELATION OF INSULIN REQUIREMENT AND RESPONSE TO CHLORPROPAMIDE THERAPY

	Previous insulin dose, units per day				Total patients	Percentage
	0	Less than 20	20–40	Greater than 40		
No. of patients	6	13	28	2	49	—
Control						
Good	6	7	7	0	20	41 }76
Fair	0	4	13	0	17	35
Unsatisfactory	0	2	8	2	12	24
Percentage satisfactory	100	85	71	0	—	—

averaged approximately one fifth that of tolbutamide, a typical experiment is shown in FIGURE 4. One patient (FIGURE 5) showing progressive resistance to tolbutamide was shifted to chlorpropamide, hyperglycemia and ketosis continued, and insulin was required for control.

Sex and race. There was no correlation between the sex or race and response to therapy (TABLE 6).

Dosage of chlorpropamide. The majority of our patients were maintained satisfactorily on doses of 500 mg. per day or less, generally given as a single

TABLE 5
COMPARISON OF TOLBUTAMIDE AND CHLORPROPAMIDE DOSAGE

	Previous maintenance dose (tolbutamide), grams per day					Total patients
	0 5	1 0	1 5	2 0	2 5	
No of patients converted	1	3	6	4	1	15
Present maintenance dose (chlorpropamide) grams per day						
0 100	1	—	2	—	—	
0 200	—	2	3	—	—	
0 250	—	—	1	1	—	
0 400	—	1	—	1	—	
0 500	—	—	—	—	1	
0 625	—	—	—	2	—	

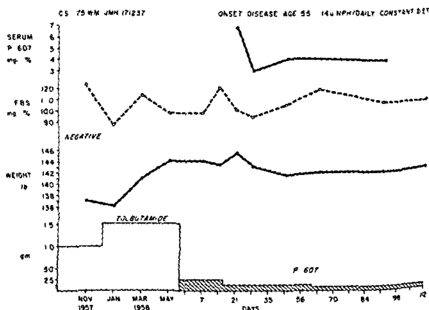


FIGURE 4 Replacement of tolbutamide by chlorpropamide The observations illustrated were made while the patient was on a constant diet in the special study unit Chlorpropamide, in a dose of 125 to 250 mg daily, maintained the satisfactory control previously achieved in this patient by 1 to 1 5 gm tolbutamide daily

dose each morning It was observed that patients taking small doses (that is 100 mg per day) could be controlled just as readily when the dose was given as a 200 mg tablet every other day (TABLE 7) In view of the relatively slow elimination of the drug it is probable that the same thing may be said of higher doses but we have no observations to support this assertion

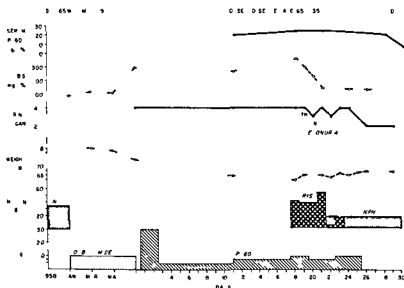


FIGURE 5 Failure of chlorpropamide in tolbutamide resistance Neither tolbutamide (1 gm daily) nor chlorpropamide (750 mg daily) produced successful control of diabetes in this patient

TABLE 6
RELATION OF SEX AND RACE TO SUCCESS OF THERAPY

	Male	Female	White	Negro
No of patients	14	35	37	12
No satisfactorily controlled	11	26	29	8
Percentage satisfactorily controlled	79	74	78	67

Diabetics not responding to 500 mg per day usually failed to respond to doses of as much as 2 gm per day At these higher dose levels toxic symptoms were not uncommon

Concentration of chlorpropamide in blood serum When chlorpropamide is administered in a constant daily dose of 1 gm or less the serum concentration of the drug rises steadily for about five days attaining a reasonably stable plateau by the end of that time The decline in serum concentration when

the drug is stopped is still slower, and this is reflected in a rather delayed escape from diabetic control in patients who cease taking the drug (FIGURE 1)

FIGURE 6a represents the range of serum chlorpropamide concentrations found in a number of patients stably maintained at various dose levels of the drug. To these data has been fitted freehand the straight line curve shown in FIGURE 6b. The assumption, which may not be entirely accurate, has been made that the relation between drug dose and serum concentration is linear. At any rate FIGURE 6a gives a rough indication of the drug concentration to be expected at any dose level.

TABLE 7
RELATION OF MINIMAL DOSE TO ADEQUATE CONTROL*

	Chlorpropamide dose (grams per day)			Total
	0 1–0 2	0 25–0 50	0 60	
No of patients	20	13	16	49
Control				
Good	14	6	—	20
Fair	6	6	5	17
Unsatisfactory	—	1	11	12

* Satisfactory control was achieved in 33 of the 49 patients (67 per cent) by a daily dose

as 2 gm daily

TABLE 8 shows that satisfactory control, if it can be attained at all, will generally be achieved at a plasma concentration of chlorpropamide of

Thirty six of the patients represented in TABLE 8 have had repeated determinations of serum chlorpropamide concentration at a stable dose level of the drug, in 3 of these, a second series of observations was made at higher dose levels and with higher serum concentrations

Of the 36 patients, 22 (61 per cent) were satisfactorily controlled at maximum serum concentrations of 15 mg or less per 100 ml. Three patients not satisfactorily controlled at the lower serum concentrations (15 mg /100 ml or less) were subsequently *given larger doses of the drug*. One remained uncontrolled with serum concentrations as high as 36 mg /100 ml, fair control was achieved in the other 2 at maximum levels of 20 and 25 mg /100 ml, respectively

Toxicity Toxic manifestations occurred in 15 of our 49 cases (30 per cent)

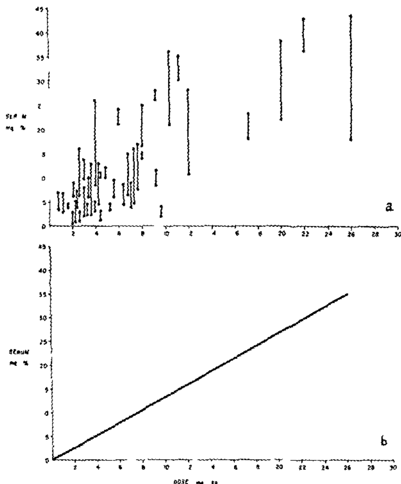


FIGURE 6 Relation of serum chlorpropamide levels to oral dosage (a) Each bar shows the range of serum chlorpropamide concentrations attained 4 days or more after any given constant daily dose of the drug had been instituted The data are based on 145 determinations in 39 patients each range shown representing on the average 3 7 determinations carried out for weeks or months at a stable dosage level (b) A free hand curve drawn to fit the data of FIGURE 6a

Most of the effects were transient and consisted of headache dizziness drowsiness itching and possible hypoglycemic symptoms (TABLE 9) There were however only 2 instances (4 per cent) where the symptoms persisted and necessitated withdrawal of the drug In 1 patient (G E) who had been taking 500 mg per day with marginal fair control the dose was increased to 750 mg per day with ensuing nausea vomiting hiccoughs and an erythem

15 mg /100 ml are reached The maximum recommended dose is generally 8 mg /kg per day (500 mg per day for the average size patient) Further increases seldom give more effective control, and they will often raise the serum chlorpropamide level to a range higher than 15 mg , where a considerable increase of toxic reactions is to be expected It is evident, however, that the dose of the drug per kilogram of body weight can influence effectiveness and toxicity only insofar as it affects the concentration of the drug in body fluids Since the data show that there is considerable variation between individuals in the serum drug concentration attained at any given dose, it would appear that the determination of serum chlorpropamide concentration will often be helpful in establishing the optimal dose more precisely and, especially, in justifying increases to more than 8 mg /kg per day in patients who, on this amount of drug, may remain uncontrolled and maintain low serum drug concentrations

The incidence of toxic reactions of some kind in our series has been high, but our figure of 30 per cent does not, of course, represent the incidence of toxicity to be expected in patients who will be treated after the principles of proper dosage have become well established In the course of preliminary, experimental clinical studies of a new drug, neither the minimum effective dose nor the incidence of toxicity at various dose levels has yet become well defined, and the majority of the toxic manifestations reported here have occurred at a higher dosage than would now ordinarily appear to be indicated

Summary

Forty nine patients with diabetes mellitus, ranging from fourteen to eighty years of age have been treated with chlorpropamide for as long as five months

Satisfactory control of the diabetes was achieved in 76 per cent of these cases, the majority of patients being those whose disease began after 40 and who required less than 40 U of insulin daily The incidence of successful control was not significantly influenced by the sex or race of the patients nor by the duration of the diabetic state

Chlorpropamide is effective in controlling diabetes, in general, under the same circumstances as tolbutamide, however, chlorpropamide is about five [illegible] on a weight basis This enhanced potency appears to [illegible] trations of [illegible] A daily [illegible] verage size [illegible] drug, but there is considerable variation, especially between individuals For this reason, serum chlorpropamide determination should prove a helpful guide to therapy

Toxic manifestations of some kind, principally mild and all reversible, occurred in 30 per cent of our cases The incidence of toxic reactions in this

series is higher than, and not representative of, that to be expected with therapeutic use of the drug which in our opinion should ordinarily consist of daily doses not in excess of 8 mg./kg. of body weight.

Acknowledgment

We are indebted to Domenic G. Iezzoni of Chas. Pfizer & Co., Inc., Brooklyn, N. Y., for supplies of chlorpropamide.

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CLINICAL AND EXPERIMENTAL STUDIES WITH CHLOR PROPAMIDE IN DIABETES MELLITUS, IN NORMAL INDIVIDUALS, AND IN NONDIABETICS WITH HEPATIC DISEASE

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The hypoglycemic action of the sulfonylureas presupposes reserves of endogenous insulin These substances probably also act through an extra pancreatic mechanism influencing intrahepatic metabolism, a process that has been confirmed clinically and experimentally by a selective accumulation of liver glycogen Because the sulfonylureas produce no increase in sugar utilization by skeletal muscle tissue, their action differs significantly from the action that is typical of exogenous insulin The liver, therefore, is one of the sites affected by sulfonylureas

It has been clinically and experimentally shown that the sulfonylureas influence intrahepatic metabolism The mode of action of chlorpropamide, *as proved in our studies, does not differ significantly from that of carbutamide* or tolbutamide In test animals the antidiabetic drugs, including chlorprop amide, produce a similar increased accumulation of glycogen in the liver In rats [illegible] (35 mg per day [illegible] exhibits [illegible] disease (hepatitis and cirrhosis), we observed a similar effect and, also an objective improvement in hepatic function, the latter was confirmed by laboratory and liver function tests, especially Bromsulphalein retention Adverse effects on hepatic function were not demonstrated in these patients nor in 35 diabetics treated with chlorpropamide orally administered for several months In rats receiving 50 mg or 100 mg /kg of chlorpropamide for 6 weeks, we observed an increase in liver glycogen, but no histopatho *logical alterations*

Because of the recent interest in the mechanism of action of the antidiabetic sulfonylurea derivates, the effect of chlorpropamide on the liver transaminase system was investigated, since an inhibition of transaminase reactions can lead to a decrease in the rate of neoglucogenesis Serum transaminase values were determined in normal individuals, hepatic patients, and diabetics during therapy Glutamate oxalacetic and glutamate pyru vate transaminase levels, as well as aldolase and lactic acid dehydrogenase activity, were investigated It was observed that chlorpropamide inhibits serum transaminase levels as do other compounds, as shown by other investi gators It is suggested that by inhibiting transamination such derivates decrease the rate of neoglucogenesis This hypothesis would also explain the hepatotropic effects

In our work at the Medical Clinic we have been particularly interested in determining minimal maintenance dosages We have under study 35

patients 25 of whom received previous treatment and 10 of whom were diagnosed during the course of the study and received chlorpropamide as the initial therapeutic agent

Six of the patients were less than 25 years old and it was in this group that we observed our 2 failures The other 4 patients are interesting since we did not observe an adequate response during the initial 2 weeks of treatment and therefore gave insulin in addition to chlorpropamide We continued this combined therapy for 14 to 21 days after which we found surprisingly that the insulin dosage could be reduced and finally discontinued

Our remaining 29 patients most of whom were 50 years of age or older and generally obese responded well to chlorpropamide It is our practice to begin treatment with 1 gm per day for 3 or 4 days and then to reduce dosage to the lowest possible therapeutic level We have found that most patients can be maintained in satisfactory balance with a dosage of 250 mg daily However we have had success with as little as 100 mg daily and in an occasional patient with 100 mg every other day

We have not observed any side effects from dosages of 1 gm daily or less On higher dosage patients may experience nausea discomfort in the epigastrium and a feeling of abdominal fullness There was no indication of clinical toxic manifestations on the blood skin or nervous systems

We were impressed with the relatively low minimal dosages of chlorpropamide that were fully effective in maintaining satisfactory sugar metabolism in patients with diabetes Tolerance of dosages of 1 gm a day or less is excellent probably more than that amount is not necessary since it may well represent the maximum therapeutic quantity The average maintenance dosage is 250 mg daily and, not infrequently even smaller doses are adequate

EXPERIENCES WITH CHLORPROPAMIDE, ESPECIALLY IN THE BRITTLE DIABETIC

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It has been shown that chlorpropamide is a sulfonylurea with antidiabetic action Early in our clinical trial of this drug, we attempted to evaluate its control of the brittle or labile diabetic We also attempted to determine whether the administration of very large doses would result in a more rapid control of the diabetes, and whether very large maintenance doses would control the brittle diabetic patient when average dosages failed to do so Thus far, 34 patients with diabetes mellitus have been given chlorpropamide under our supervision All of these patients were adhering to a restrictive diet of the American Diabetic Association type, 11 were taking, in conjunction with the diet more than 34 U of insulin daily in order to achieve control of their diabetes

Of the 34 diabetic patients receiving chlorpropamide therapy, the drug was discontinued in 5 because of ineffectiveness of control, and in 5 others because of untoward effects One patient was lost to our control, and 1 patient died

Initial Dosage

Twenty one patients were given an initial dosage of more than 1 gm of chlorpropamide daily Of these, 12 achieved adequate control Two patients received 4 gm as an initial dosage, neither achieved adequate control Ten received 3 gm as an initial dosage, of these, 6 were inadequately controlled Five patients received initial dosages of 2 gm, 3 of these were inadequately controlled (TABLE 1)

TABLE 1
CONTROL ACHIEVED WITH MORE THAN 2 0 GRAMS OF CHLORPROPAMIDE AS INITIAL DOSE

Initial dose (gm)	Number of patients	Control	
		Adequate	Inadequate
2 0	5	3	2
3 0	10	4	6
4 0	2	0	2

Maintenance

Seven of our patients received maintenance dosages of more than 1 gm daily, of these, 3 had adequate control Of 3 patients receiving more than 2 gm and as much as 4 gm daily for maintenance, none was controlled

One of these patients received 4 gm daily for 4 days and achieved a blood level of chlorpropamide of more than 50 mg per cent Although she had no toxic effects or symptoms from this drug, it was discontinued because of lack of control (TABLE 2)

TABLE 2
MAINTENANCE DOSE (CHLORPROPAMIDE ONLY) AND RESULTS ACHIEVED

Maintenance dose (gm)	Number of cases	Control	
		Adequate	Inadequate
0 25	1	1	
0 25–0 5	14	12	2
0 6–1 0	12	6	6
1	4	3	1
2 3	0	0	0
3	0	0	0
4	3	0	3

Control

Of 11 patients requiring a restrictive diet and more than 34 U of insulin daily for control of their diabetes, only 4 were able to continue chlorpropamide therapy for more than 4 weeks The other 7 were returned to insulin because of inadequate control of these, 6 had disturbing side effects that, in themselves would have been sufficient reason for discontinuing the drug Three patients in whom control of the diabetes was lost on tolbutamide therapy were well controlled on chlorpropamide

Side Effects

The number of side effects seemed to bear no direct relationship to the maintenance dosage of chlorpropamide More side effects were noted in

TABLE 3
CORRELATION OF DAILY DOSAGE OF CHLORPROPAMIDE WITH SIDE EFFECTS

Dosage (gm)	Number of cases	Side effects			
		Skin	Gastrointestinal	CNS	Other
0 25	1				
0 50	14	2	5	6	
1 0	12		3	3	
3 0	7				1*
Total		2	8	9	1*

* Deceased

patients receiving 0 5 gm daily than in those on higher daily dosages (TABLE 3) One patient, who had received large doses of chlorpropamide, died while receiving the drug He suffered from arteriosclerotic and coronary artery heart disease in association with probable portal cirrhosis of the liver Despite symptoms of anorexia and lethargy, the drug was continued until about thirty six hours before death At no time was hypoglycemia present Autopsy findings showed nonspecific changes of fatty metamorphosis of the liver, cerebral atrophy, an old healed myocardial infarction, and slight aortic atherosclerosis

Conclusions

In a small series of thirty four cases studied thus far we have found that

Chlorpropamide was not effective in control of the brittle diabetic

The effectiveness of chlorpropamide is not increased by large dosages, even as much as 4 gm daily

The toxicity of this drug apparently increases with large doses, but some patients can tolerate large daily doses and high blood levels without ill effects

High dosage of more than 1 0 gm daily was not associated with a higher incidence of side effects

The drug is apparently effective in the mild diabetic

Three patients not controlled by tolbutamide were controlled adequately by chlorpropamide

One patient died while on large doses of chlorpropamide The exact cause of death is unknown

A PRELIMINARY METABOLIC AND CLINICAL EVALUATION OF CHLORPROPAMIDE

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The metabolic effects and clinical efficacy of chlorpropamide, an oral hypoglycemic agent with potential therapeutic value, were evaluated in twenty three patients with diabetes mellitus A preliminary report of this experience with chlorpropamide is presented here

Materials and Methods

During the course of these studies 10 patients with diabetes mellitus were selected for special study on the metabolic ward Four of these patients, when placed under conditions of strict diet control similar to that previously recommended for them, did not demonstrate glycosuria and required no insulin Accordingly trials of chlorpropamide therapy were not made with these 4 patients, and they were discharged from the hospital with diet therapy alone The other 6 were considered suitable for the metabolic studies described below Brief descriptions of these patients are included with the data obtained for each, as presented under *Results*

The patients were maintained on constant diabetic diets designed to meet individual requirements, each daily diet being divided into 4 feedings The patients were weighed daily fluid intakes were recorded, but the subjects were permitted to drink water at liberty Twenty four hour outputs of urine were collected and preserved with 10 ml of a 1 10 mixture of toluol and glacial acetic acid and refrigeration Each daily urine volume was analyzed quantitatively for sugar content [1, 2]

The periods of study ranged from 18 to 62 days (average 39 days) Control periods of from 3 to 8 days preceded the administration of chlorpropamide in all patients When it seemed permissible, insulin was withdrawn or reduced in dosage before the drug was given The chlorpropamide* was given orally, 1 gm daily each morning before breakfast One-gram doses of tolbutamide† were similarly given Tolbutamide in daily doses greater than 1 gm was administered in divided doses at mealtime

Fasting blood samples were obtained frequently for sugar determinations [1, 2] Fasting blood levels of chlorpropamide and of tolbutamide were determined serially in some patients [3] For 3 patients, blood levels of chlorpropamide were determined several times within the 4 hour period immediately following a 1-gm oral dose of the drug

These studies included frequent urinalyses, complete blood counts, and tests of liver functions, including serum bilirubin concentration, alkaline phosphatase, cephalin cholesterol flocculation, and thymol turbidity In

* Provided by Chas Pfizer & Co, Inc Brooklyn, New York

† Provided by The Upjohn Company, Kalamazoo, Mich

some patients Bromsulphalein excretion tests also were obtained before during and after the administration of the hypoglycemic agent In addition the patients were observed closely for evidences of untoward reactions to chlorpropamide and for symptomatic hypoglycemia

Chlorpropamide was also evaluated in 3 patients with diabetes, who were hospitalized on the general medical wards and in 14 patients attending the diabetes clinic as outpatients These patients were selected on the basis of their likelihood to respond to an oral hypoglycemic agent for example patients with stable maturity onset type diabetes The 3 hospitalized patients had not received insulin for at least 3 days before chlorpropamide was given They were given 1 gm chlorpropamide daily for at least 6 days The outpatients were instructed to discontinue insulin and to begin chlorpropamide on the same day Initial dose of chlorpropamide for the outpatients was 1 gm daily for 1 week and 0 5 gm daily was prescribed thereafter This therapy was continued for from 1 to 3 months (average 9 weeks) The patients visited the outpatient department weekly No special effort was made to ensure more adequate adherence of the patients to their diets or to other diabetic instruction previously given They were advised to continue their diabetic regimens as in the past These patients were aware of course that a change in medication was made for example an injection of insulin was being replaced by an oral medication and in some cases, the frequency of required visits to the outpatient department was increased Chlorpropamide was evaluated in the outpatients by fasting blood sugar determinations and fractional qualitative urine sugar determinations (Somogyi method) Blood sugar determinations were done almost daily in the hospitalized patients and usually weekly in the outpatients In addition daily quantitative glycosuria was determined for the patients in the hospital Blood chlorpropamide levels were determined at weekly intervals in some of the outpatients Urinalyses hemograms blood urea nitrogen concentration and liver function tests were also done at intervals for these patients

Results

Metabolic studies Since it was not possible to carry out identical protocols for the six patients studied on the metabolic ward the data for each of these patients are presented separately

Patient J M a 42 year old Negro male diabetic for 1 year was admitted to the hospital because of diabetic acidosis Of his own accord he had stopped taking insulin 6 months prior to admission The acidosis was treated and he required 15 U NPH insulin daily for 2 weeks preceding the metabolic study The results of this study are presented in FIGURE 1 During the initial 7-day period of metabolic study this patient's diabetes was well controlled with a 2500 cal diet and 15 U of NPH insulin daily Insulin was then discontinued During the 6 days without insulin glycosuria increased from less than 5 gm to about 30 gm daily and fasting blood sugar levels increased from 160 mg to 210 mg per cent Then 1 gm chlorpropamide was given orally daily In association with chlorpropamide therapy glycosuria and fasting blood sugar levels promptly decreased

Twenty four hours after the initial 1 gm dose had been given fasting blood sugar level had decreased from 210 mg to 164 mg per cent and, after several days ranged between 110 and 132 mg per cent Glycosuria was less than 5 gm daily and remained so even after the dose of chlorpropamide was decreased to 0 5 gm daily on the tenth day of therapy Upon discharge from the hospital a similar diet and 0 5 gm chlorpropamide were prescribed for this patient who has been seen in the outpatient clinic at regular intervals and has maintained excellent control of diabetes under this regimen during

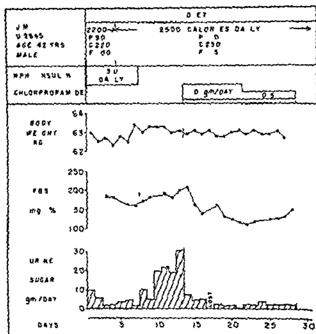


FIGURE 1

the 6-month period to date The patient demonstrated a prompt and adequate hypoglycemic response to chlorpropamide administered orally and it replaced insulin in his diabetic regimen

Patient A M a 60 year old Negro female with stable easily controlled diabetes of 10 years duration required 30 U of NPH insulin daily This patient had a past history of repeated episodes of pancreatitis and it seemed probable that the diabetes was secondary to chronic pancreatitis She was being treated in the hospital for pulmonary tuberculosis and received isoniazid and para aminosalicylate while this study was in progress Insulin was discontinued 1 week before metabolic study was initiated The findings in patient A M are presented in FIGURE 2 During the first 3 days of study this patient was given no insulin but was maintained on a constant 2000-cal

diabetic diet Glycosuria increased from 68 gm to 94 gm daily, fasting blood sugar levels increased from 194 mg to 360 mg per cent, and the patient lost weight Chlorpropamide, 1 gm daily, was then given orally After 48 hours, glycosuria had decreased to 54 gm daily, fasting blood sugar level had decreased to 228 mg per cent, and body weight was maintained Chlorpropamide was given for 6 days, following which fasting blood sugar levels

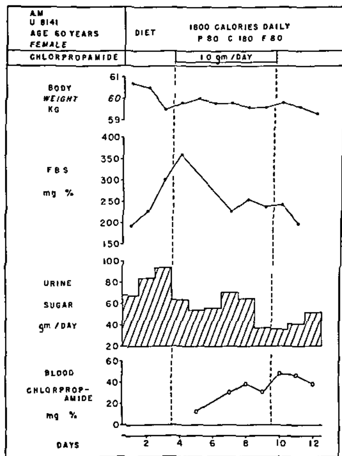


FIGURE 2

were about 200 mg per cent and glycosuria 35 to 40 gm daily. Although this patient demonstrated a hypoglycemic response to chlorpropamide, glycosuria was not decreased sufficiently to warrant considering diabetic control as excellent Fasting blood levels of chlorpropamide were determined in this patient during and after the administration of the drug These levels increased progressively during therapy to 54 mg per cent (FIGURES 2, 3)

Patient M L, a 62 year old Negro female, had stable diabetes of 12 years' duration that was not well controlled with 10 U NPH insulin daily She

was admitted to hospital for regulation of diabetes This patient also had arteriosclerotic heart disease and took digitalis daily The initial 5 day control regimen for M L (FIGURE 4) consisted of a 2000 cal diet and 10 U of NPH insulin daily During this period fasting blood sugar levels ranged ... Beginning ... to the ... ia and ... ly and ... liscon ... which

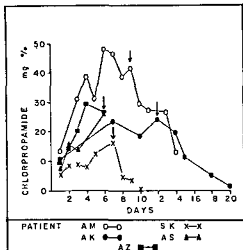


FIGURE 3 Blood levels after oral administration of 1 gm per day of chlorpropamide Arrows indicate the days on which chlorpropamide was discontinued

eventual replacement of insulin by this agent The apparent slow response to chlorpropamide makes this study somewhat difficult to interpret but since it had been known that this patient had diabetes for 12 years and had required insulin during this time this case is considered a positive response to chlorpropamide

Patient A K (FIGURE 5) a white female 63 years of age was found to have diabetes following a recent admission to the hospital for thrombophlebitis of the left lower leg She also demonstrated senile dementia but was able to cooperate in the adequate collection of urine specimens Prior to the initiation of study she had persisting glycosuria of about 20 gm daily although she was given a prescribed diet and 40 U of NPH insulin daily

This patient had about 25 gm glycosuria daily, and fasting blood sugar levels of 280 mg per cent during the 2 initial days of observation on the metabolic ward while she was consuming a 2000 cal diet and receiving 40 U of NPH insulin daily. Two grams of tolbutamide, given orally, in addition to the insulin, produced a prompt decrease in glycosuria to less than 5 gm daily and in fasting blood sugar to 170 mg per cent, this was apparent after the first day of therapy. Discontinuance of insulin was followed by transitory increases in urine and blood sugar levels, but control of diabetes continued

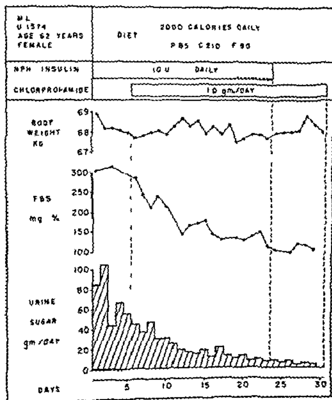


FIGURE 4

to be adequate. After 11 days of tolbutamide administration, this agent was discontinued following which glycosuria and hyperglycemia increased, reaching values of 30 gm daily and 250 to 280 mg per cent ... after 8 days. One gram of *chlorpropamide* ... 13 days. Chlorpropamide ... glycem...

... was not decreased ... as with tolbutamide. After chlor- a

propamide was discontinued glycosuria and hyperglycemia increased during a 9-day control period During a final period of study 1 gm of tolbutamide was given daily As in the earlier period of tolbutamide administration the effect of this agent on blood sugar and glycosuria was prompt The urine sugar excretion decreased from about 30 gm daily to less than 10 gm on the first day this drug was used Maximal blood sugar response was noted 48 hours after tolbutamide therapy was started by which time levels

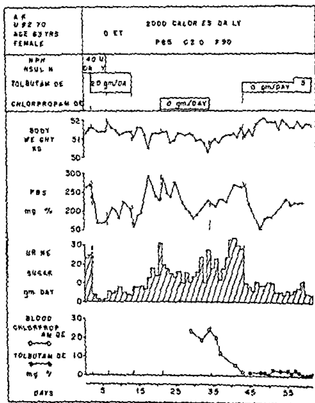


FIGURE 5

had decreased from about 265 mg per cent to 154 mg per cent Blood sugar then increased gradually to approximately 230 mg per cent at which level it became stable glycosuria averaged less than 10 gm daily Increasing the dose of tolbutamide from 1 0 to 1 5 gm had no appreciable effect on blood or urine sugar levels

Serial fasting blood levels of chlorpropamide and tolbutamide were done for this patient (FIGURE 5) During the last 6 days of chlorpropamide therapy blood levels of this agent ranged from 18 to 25 mg per cent After

chlorpropamide was disco
blood Approximately 7
Only small amounts of t
samples obtained during the administration of this agent

Both chlorpropamide and tolbutamide demonstrated hypoglycemic and aglycosuric effects in patient A K The response to tolbutamide was more prompt and more striking than the response to chlorpropamide

Two additional patients (E H and S K) who were studied on the metabolic ward for thirty six and sixty days, respectively, failed to demonstrate any significant responses to the oral administration of chlorpropamide or of tolbutamide Brief résumes of these patients follow

Patient E H, a 46 year old man, had very labile diabetes He had experienced frequent episodes of acidosis and hypoglycemia, many of which required hospitalization during the 10 years since the diagnosis of diabetes mellitus was made His usual daily insulin dosage was 40 U of PZI insulin each morning and 30 to 50 U of regular insulin during the day He was not well controlled despite many efforts in the hospital to accomplish an adequate regimen His basal control regimen consisted of a 1900 cal diet and insulin (as indicated above) Following a 12 day control period, he was given, in addition, 1 gm chlorpropamide daily for 22 days This period of chlorpropamide administration was followed by the basal regimen for 14 days Then 2 gm tolbutamide was given orally each day for 13 days There was no effect attributable to either of these hypoglycemic agents

Patient S K, a lean man 47 years of age, was admitted to hospital in
He responded promptly to
controlled
ays in hos
pital, he was transferred from a general medical ward to the metabolic ward

glycemia or urinary sugar excretion Following a 3 day control period without insulin tolbutamide was given orally (1 gm , 3 gm , and 4 gm orally daily for 3 successive periods of 3 days each) Tolbutamide was also without

d
n
6 to 17 mg per cent After tolbutamide had been given for 9 days in dosages increasing from 1 gm to 4 gm daily, 14 mg per cent of tolbutamide was detectable in fasting blood specimens The failures of response in this patient could not therefore be attributable to a failure of absorption of the hypoglycemic agents In S K, approximately 3 days were required for

the complete disappearance of chlorpropamide from the blood after cessation of therapy

Chlorpropamide blood levels In some patients, chlorpropamide blood levels were determined at intervals during the 4 hour period following a 1 gm oral dose These data are presented in FIGURE 6 The average blood level was about 9 mg per cent after 1 hour, and 12 mg per cent 4 hours after its ingestion, indicating prompt absorption of this agent

The serial chlorpropamide blood levels obtained during the studies of patients on the metabolic ward are presented compositely in FIGURE 3 The patients received 1 gram of chlorpropamide daily for the time indicated

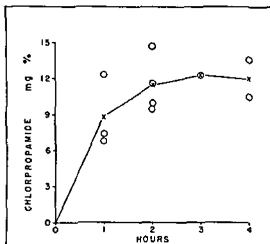


FIGURE 6 Blood levels after oral administration of 1 gm chlorpropamide Curve from ╲ to ╲ indicates the average value for each interval following chlorpropamide administration

on the chart Blood levels of the drug were cumulative at this dosage This relatively small group of patients demonstrated considerable variation in blood level attained with comparable doses of the drug The rates of disappearance of chlorpropamide from the blood after therapy varied from 3 to 7 days in these patients

The blood levels of chlorpropamide determined at weekly intervals in some of the patients attending the outpatient clinic are presented in TABLE 1 Although these data are admittedly meager, they suggest that maintenance doses of 0 5 gm daily of chlorpropamide may be adequate to maintain blood

60 year old female newly discovered diabetic) responded adequately to chlorpropamide, in a dosage of 1 gm daily for 6 days with a decrease in fasting blood sugar to levels below 150 mg per cent, she became essentially

aglycosuric Continued on a maintenance dose of 0 5 gm daily she has been observed in the outpatient clinic for 2 months during which time diabetic control has been sustained The other 2 patients of this group (a 70 year old female and a 46 year old male) each of whom required 20 U of NPH insulin for control of diabetes did not respond adequately to 1 gm chlorpropamide given daily for 1 week they were returned to insulin therapy

The fourteen outpatients selected for study ranged in age from thirty to eighty three years with an average age of sixty one years Diabetes had been established for periods of from six months to twenty eight years The average duration of diabetes was ten years In all patients the diabetes was the stable maturity onset type

TABLE 1
CHLORPROPAMIDE BLOOD LEVELS IN DIABETIC OUTPATIENTS

Weeks of therapy	Daily dose (gm)	No of patients	Level mg per cent	
			Range	Average
1	1 0	3	30–34	32
3	0 5	6	14–33	23
5	0 5	3	13–24	18
7	0 5	2	18–20	19
9	0 5	2	20–23	22

Three of the patients had been treated with diet therapy only the remainder had been treated with diet and from *10 to 35 U insulin The average* insulin dose was 25 U daily

All patients were given similar doses of chlorpropamide 1 gm per day for the first week followed by one half gm per day as a maintenance dose *thereafter*

An adequate clinical response to this drug was established as one exhibiting aglycosuric fractional urines lack of subjective symptoms maintenance of body weight and reduction in fasting blood sugars—preferably below 150 mg /100 cc

In 12 patients the diabetes was adequately controlled with a single daily dose of chlorpropamide as judged by the above criteria At present these patients continue to be treated with diet and chlorpropamide therapy Seven showed an average reduction in the fasting blood sugar of 105 mg per cent from previous levels and continued to be maintained at levels below 150 mg per cent Seven others showed no appreciable change in fasting blood sugar levels however most of them had been well controlled on their pr [illegible] therapy

[illegible] One [illegible] viously [illegible] *atment* controlled with 15 U [illegible]

and was returned to insulin therapy This patient was the only one to express subjective symptoms to chlorpropamide therapy She complained of epigastric distress during treatment, which disappeared when chlorpropamide was discontinued, she was being treated simultaneously for a chronic duodenal ulcer

The other patient, a 45 year old male with mild diabetes of 3 years' duration, previously controlled with 25 U of insulin, did not respond adequately after 4 weeks of treatment with chlorpropamide, insulin therapy was resumed

Chlorpropamide appeared successful in controlling the diabetes in 12 of 14 of this group of selected outpatients, most of whom were in the older age group with stable diabetes previously treated with diet therapy alone or in conjunction with moderate doses of insulin Control has been maintained for as long as 6 months

Toxicity With the exception of the patient mentioned above, who experienced epigastric distress, there were no subjective symptoms of toxicity nor were there laboratory evidences of disturbances of renal, hepatic, or hematological function in any of the patients studied Symptomatic hypoglycemia did not occur

Discussion

Chlorpropamide was an effective oral hypoglycemic agent when administered orally to most of the patients (with diabetes mellitus) studied As with other oral hypoglycemic agents patients with stable, maturity-onset type diabetes demonstrated more adequate responses than relatively young patients with labile diabetes There were exceptions to this, however, in that two of the outpatients studied who were judged to fulfill the accepted indications for therapy with an oral agent, failed to respond to chlorpropamide Furthermore, the patterns of response to chlorpropamide, as evaluated in the six patients studied on the metabolic ward, were quite variable One patient demonstrated a prompt and adequate response (J M, FIGURE 1), another, a delayed response (M L, FIGURE 4) others showed significant but inadequate responses (A M, FIGURE 2, A K, FIGURE 5) The reasons for this variation are not apparent they did not relate to the blood level of chlorpropamide Although blood levels varied considerably among patients given similar doses of the drug (FIGURE 3), blood level did not correlate with the adequacy of diabetic control attained with chlorpropamide

Four patients who either failed to respond or demonstrated inadequate responses to chlorpropamide were subsequently given tolbutamide Three of these also failed to demonstrate responses to tolbutamide (E H, S K, A M) A fourth patient (A K, FIGURE 5) demonstrated more prompt and pronounced effects on blood and urine sugar when she was given tolbutamide than when she was given chlorpropamide in comparable dosage

Studies of the level of chlorpropamide in the blood following its oral administration indicated that it is promptly absorbed and slowly excreted Three to seven days were required after cessation of therapy before the drug disappeared from the blood In this respect it differs from tolbutamide, which is promptly absorbed and more rapidly removed from the blood With 1 gm

daily doses of chlorpropamide blood levels of the drug progressively increased in some patients Those who demonstrated adequate responses to 1 0 gm of chlorpropamide given daily for about 1 week subsequently sustained adequate control of diabetes with a continuing dose of 0 5 gm daily Pre

daily (FIGURE

mg per cent

possible that doses less than 0 5 gm daily might have been satisfactory for maintenance in some patients On the other hand some patients might have demonstrated more adequate responses had larger doses been employed (S K FIGURE 3) In the studies reported here no effort was directed to establish either the minimally effective dose of chlorpropamide or the maximum safe dose

An important consideration in the acceptance of a drug for clinical use is its freedom from untoward effects Although no serious untoward symptoms or laboratory findings resulted from the administration of chlorpropamide to the twenty three patients studied, conclusions regarding toxicity in man must be based on a much larger experience than is reported here

Summary

The clinical and metabolic effects of chlorpropamide administered orally were evaluated in 23 patients with diabetes mellitus Six of these were studied under carefully controlled conditions on a metabolic ward The *other 17 were observed on the general medical wards (3 patients) or in the* outpatient clinic (14 patients)

Chlorpropamide given orally (1 gm daily) decreased glycosuria and fasting *blood sugar levels in 4 of the 6 patients studied on the metabolic ward* Adequate clinical responses were obtained in 2 stable diabetics for whom insulin was completely replaced Inadequate responses were found in *2 other stable diabetics who had required 40 U of NPH insulin daily* One of the patients who was inadequately controlled with chlorpropamide demonstrated more prompt and adequate decreases in glycosuria following identical doses of tolbutamide The other 2 patients (a lean 47 year old man with newly discovered diabetes and a 46 year old man with very labile diabetes)

tolbutamide

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the blood following cessation of therapy Blood levels attained with the same dose (1 gm daily) of chlorpropamide demonstrated considerable variation in the patients studied

ed in 17 patients who were

6 months with a single daily oral dose of chlorpropam

1 week and then 0 5 gm daily) With this dosage schedule, chlorpropamide blood levels of about 20 mg per cent were maintained

With the exception of epigastric distress in one patient there were no

untoward clinical manifestations attributable to chlorpropamide Symptomatic hypoglycemia was not observed Serial urinalyses hemograms and tests of hepatic and renal functions were not altered during the periods of study

Acknowledgment

The chlorpropamide and tolbutamide blood levels were determined by arrangement with Max Miller, Western Reserve University School of Medicine These determinations were done in his laboratories by Naomi F Nickerson The authors are grateful for this contribution to the study

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CHLORPROPAMIDE A NEW SULFONYLUREA STUDIED IN THE TREATMENT OF THIRTY DIABETIC PATIENTS

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A few years ago insulin was the only drug available for the control of diabetes In 1955, German authors[1 2] reported the results of the treatment of diabetic patients with carbutamide, an oral sulfa drug with a marked hypoglycemic effect Shortly thereafter a new compound known as tolbutamide, closely related to the first, was also employed[3] Loubatieres reviewed in a complete and concise presentation the history and development of these compounds[4]

[illegible]

diabetics or in thin mature individuals who have received insulin for several years

Chlorpropamide, a recently developed oral drug closely related to tolbutamide has also shown a hypoglycemic effect in animal studies The purpose of this paper is to report our experience during the last eight months in administering this compound to diabetic patients

Methods

[illegible] and 26 males) observed

20 as moderately severe and 6 as mild The disease had started from a few months to 20 years before but in most cases, it had existed 1 to 8 years There was 1 case of diabetes (No 29) due to tumoral invasion of the pancreas

Twenty four patients were on insulin therapy or had received this drug for some time Twenty four were taking or had taken oral drugs 12, carbutamide and 12 tolbutamide Five had not had any previous therapy [illegible]

Table 1 contains all pertinent data concerning each of the patients studied

A careful physical examination was performed blood sugar[5] was determined at least twice a week, and urine was examined for sugar four times a day before meals The following tests were also done complete urinalysis, blood count and differential, blood urea creatinine and bilirubin, thymol turbidity colloidal gold, cephalin cholesterol flocculation, serology and sedimentation rate

The patients were on a standard diabetic diet Oral antidiabetics were

discontinued for 1 or 2 weeks and insulin injections were discontinued for 48 hours A chlorpropamide tolerance test was then performed A fasting venous blood sample having been obtained 3 gm of the drug was given orally 4 more hourly blood specimens were then taken Blood sugar was determined according to Folin's micromethod

TABLE 1
DATA ON THIRTY PATIENTS TREATED WITH CHLORPROPAMIDE

Case no	Age years	Sex	Weight per cent	Severity of diabetes*	Duration years	Insulin	Carbutamide	Tolbutamide	Chlorpropamide tolerance test First hour	Second hour	Fourth hour	Blood sugar reduction percentages	Average doses per day	Length of treatment days	Results‡
1	56	M	−3	+	1	0	0	0					0 125	90	E
2	42	F	+38	++	4	+	0	+	190	145	90	53	1	7	G
3	51	F	+38	++	7	+	+	+	185	185	134	28	1 250	10	B†
4	55	M	+12	++	7	+	0	0	178	155	123	31	0 750	98	E
5	49	M	+22	++	6	+	+	0	95	100	88	8	0 500	180	E
6	53	M	+11	+	1	0	0	0	356	270	238	18	0 250	150	E
7	44	M	+15	+	4	+	0	0	107	89	86	20	0 250	105	E
8	17	M	−6	+	1	+	0	0	80	50	30	63	1	105	E
9	55	M	−28	+++	12	+	0	+	250	280	290	+4			
10	68	M	−5	++	18		+	0	130	83	57	56	0 250	197	E
11	40	M	+19	++	4	+	+	0					0 875	119	E
12	21	M	13	+++	4	+	0	0	215	215	215	0			
13	63	M	+11	++	8	+	0	+	180	160	148	18	1 500	63	F
14	46	F	+5	++	3	+	0	+	160	150	129	20	1	28	G
15	65	M	−14	++	8	+	0	+	300	320	295	2			
16	31	M	+8	+	1	0	0	0	228	198	170	26	0 250	111	E
17	53	M	+16	++	4	+	+	+	160	125	93	42	0 500	56	E
18	70	M	+19	++	10	+	+	0					0 250	191	E
19	57	M	+7	++	2	+	0	+	142	85	95	33	0 250	185	E
20	67	M	−20	++	8	+	+	0					0 125	200	E
21	57	M	−14	++	1	+	+	+	222	216	178	20	2	30	E
22	32	F	+19	++	10	+	+	0	190	190	120	37	1 500	213	G
23	46	M	+32	++	9	+	+	0	128	120	110	14	0 750	200	E
24	42	M	−1	+++	5	+	0	+	225	240	230	+2			
25	35	M	−11	+	1	0	0	0	170	152	112	34	0 500	119	E
26	65	M	−24	++	20	+	+	+	169	107	86	49	1 250	70	G
27	54	M	−5	+++	5	+	0	0	175	210	215	+23			
28	52	M	+18	++	1	0	0	0	194	170	116	40	0 500	10	E
29		M			6	+	+	0	180	155	140	22	2 250	22	G
30	43	M	+25	++	9	+	0	+	230	187	160	31	1	8	F

* Key + mild ++ moderately severe +++ severe
† Treatment discontinued because of intolerance
‡ Key E excellent G good F fair B bad

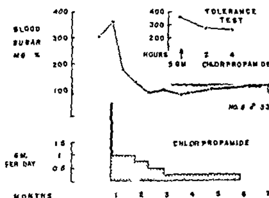
This initial test dose was followed in most of the cases by 1 gm daily taken one half after breakfast and one half after dinner This dosage was gradually reduced according to the blood sugar level Control was continued every 2 or 3 weeks at the outpatient clinic Sugar was checked in blood and

urine each time Periodically, hepatic, renal, and hematological tests also were performed

Results

Twenty four of 30 patients studied have been adequately controlled with chlorpropamide and a standard diabetic diet for as long as 7 months. Fourteen had a blood sugar reduction of 25 per cent or more when the tolerance test was performed FIGURE 1 illustrates a response typical of patients in the group Four patients had reductions of 20 per cent, and 1 each of 18, 14, and 8 per cent The average maintenance dose was 600 mg per day, varying between 125 and 2000 mg

One patient (No 21), who had a 20 per cent blood sugar reduction at the tolerance test, could not be controlled with chlorpropamide alone and



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required insulin Nevertheless after several days of combined therapy, it was found that insulin could be discontinued leaving the patient on the oral drug alone (FIGURE 2) It is interesting to note that this patient had also had a 21 per cent blood sugar reduction when 3 gm of tolbutamide was given however, control could not be achieved with this drug alone

Another patient (No 13) had an 18 per cent sugar reduction, but only a partial control was obtained Symptoms disappeared, but blood sugar level was brought no lower than 150 mg per cent

The tolerance test was negative in five patients (Nos 9, 12, 15, 24, and 27), showing either no reduction of the initial blood sugar level or a rise All had shown the same lack of response to carbutamide and tolbutamide and all had a diabetes of long duration and had been on insulin therapy for more than four years

Chlorpropamide, 1 5 gm daily, was given in conjunction with insulin in 3 of these cases A mild favorable effect was demonstrated in 2 (Nos 12 and 15) and none in the third (No 27)

Chlorpropamide was tolerated very well by most of the patients There were two (Nos 13 and 20) who complained of mild dizziness (like drunkenness) not related to hypoglycemia and lasting a few days During this time they were able to continue their work The discomfort gradually disappeared and the treatment was not discontinued

Only 1 patient (No 3) showed marked intolerance She had had a 28 per cent blood sugar reduction on the initial test and did not complain of any immediate discomfort at that time Nevertheless she developed nausea

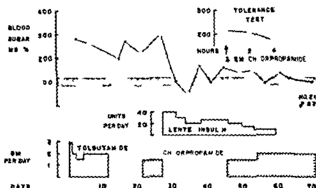


FIGURE 2 Chart of a 57 year-old Negro male (patient No 21) who had a 21 per cent reduction in blood sugar during the tolbutamide tolerance test and a 20 per cent reduction with chlorpropamide Blood sugar could not be brought to normal after 12 days treatment with tolbutamide (2 gm per day) Chlorpropamide (1 5 gm per day) was given for 8 days without success Forty U per day of lente insulin was then injected blood sugar was lowered rapidly to normal levels After 17 days of good carbohydrate metabolism chlorpropamide was resumed and insulin was gradually reduced and finally discontinued

vomiting epigastric burn diarrhea dizziness and headache after a few days of treatment at a dosage of 1 5 gm daily Symptoms stopped when the drug was discontinued but reappeared each time an attempt was made to resume the treatment She had tolerated tolbutamide well but the drug did not control her diabetes

In none of the thirty cases were skin reactions or hematological renal or hepatic changes observed

No particular reaction was noticed in four patients (Nos 5 19 20, and 23) who had pulmonary tuberculosis One with a partial gastrectomy performed 3 years before because of a perforated duodenal ulcer had an excellent evolution He is taking 250 mg every other day and has been observed for 200 days

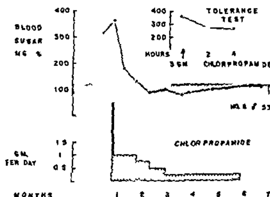
No changes in the blood pressure level occurred Two patients (Nos 6 and 17) with high blood pressure and normal renal function tests had to continue taking hypotensive drugs Two others with arteriosclerosis and high blood pressure did not show any change (Nos 10 and 26) One (No 10) who had renal insufficiency (urea 100 mg per cent) was maintained on a low protein diet during 197 days of therapy In addition to an excellent carbo-

urine each time Periodically hepatic, renal, and hematological tests also were performed

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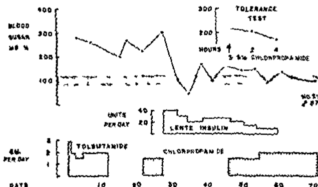


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No particular reaction was noticed in four patients (Nos 5 19, 20, and 23) who had pulmonary tuberculosis One, with a partial gastrectomy performed 3 years before because of a perforated duodenal ulcer, had an excellent evolution He is taking 250 mg every other day and has been observed for 200 days

No changes in the blood pressure level occurred Two patients (Nos 6 and 17) with high blood pressure and normal renal function tests had to continue taking hypotensive drugs Two others with arteriosclerosis and high blood pressure did not show any change (Nos 10 and 26) One (No 10), who had renal insufficiency (urea, 100 mg per cent) was maintained on a low protein diet during 197 days of therapy In addition to an excellent carbo-

hydrate balance, a slight decrease in blood urea level was achieved (urea, 80 mg per cent) This patient is now taking 250 mg per day

Discussion

Chlorpropamide seems to have the same mode of action as carbutamide and tolbutamide, but it seems to be more powerful most patients requiring a lower dosage

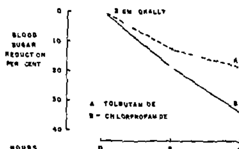


FIGURE 3 Comparative average blood sugar reduction (percentage) in nine patients Reduction was 17 5 per cent for tolbutamide and 35 3 per cent for chlorpropamide

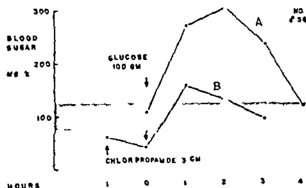


FIGURE 4 An abnormal oral 100 gm glucose tolerance test (patient No 1) became almost normal when repeated 1 hour after administration of 3 gm of chlorpropamide

When a 4 hour tolbutamide tolerance test was performed on 9 patients, an average blood sugar reduction of 17 5 per cent was found A chlorpropamide

propamide previously had not been balanced with other oral drugs alone, requiring insulin also

An abnormal glucose tolerance test in 1 patient (No 1) showed an almost

normal reaction when repeated 1 hour after the patient had taken 3 gm of chlorpropamide (FIGURE 4)

It is interesting to note that a seventeen year old patient (No 8) has been very well controlled with this drug

Summary

The activity of chlorpropamide in 30 diabetic patients formerly treated with insulin carbutamide, or tolbutamide has been studied Venous blood sugar levels during the 4 hours following oral administration of 3 gm of chlorpropamide were previously determined The drug was administered to 27 of these patients for as long as 7 months The percentage of blood sugar reduction obtained in the initial test is related to the results of the treatment The curves plotted from these data are compared with similar tests that used tolbutamide

The drug was well tolerated Its mode of action seems to be similar to that of previously used sulfa drugs Nevertheless chlorpropamide seems to be more powerful and, in some cases, more effective, a lower dosage was required by most patients It was possible to keep in good balance some cases that had not responded adequately to other oral drugs No skin reactions and no hematological, renal, or hepatic changes were observed

Acknowledgment

The authors are indebted to German Stiglich and Luis Espejo for their cooperation The valuable technical assistance of Laura Garcia and Albuina Muñoz is acknowledged The chlorpropamide used was furnished by Chas Pfizer & Co , Inc , Brooklyn, N Y

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PRELIMINARY REPORT ON THE TREATMENT OF DIABETES MELLITUS WITH CHLORPROPAMIDE*

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Chlorpropamide was made available to us early in July 1958 To initiate the investigation, 113 unselected diabetic patients were chosen Twenty-five of these were rejected because they showed satisfactory control with diet alone, and 6 were considered not suitable for exposure to chlorpropamide because of severe metabolic nephropathy In 3 cases treatment was suspended because of intercurrent conditions not related to the use of the drug under investigation and, in 27 cases, the study was discontinued for non-medical reasons Of the original group, 52 were left to be considered in this study which, because of the small number of cases that have been studied thus far and the shortness of the observation periods (5 to 36 days), should be regarded as a preliminary report

Material and Methods

All patients were treated on an ambulatory basis, they and their relatives received careful instructions regarding such aspects as diet and the performance of urine test for sugar On the days the blood sugar was determined the patients were admitted to the outpatient dining room of the Monte Carmelo Medical Clinic, where the meals were served Every patient under study was submitted to the following controls (1) medical consultation at least once a week, (2) determination of venous blood sugar (Folin Wu method), while fasting and between meals, (3) routine urinalysis, and (4) hematological studies (red and white cells, platelet counts, hemoglobin and hematocrit determinations, and differential white counts), as well as blood urea nitrogen determinations

Generally speaking, the diets prescribed had the following approximate composition for a 24 hour period 200 to 250 gm of carbohydrates, 1 to 1 5 gm of protein per kilogram of body weight, and fat to complete the caloric requirements Because of food habits in Venezuela, the diet contained liberal amounts of foods rich in easily available carbohydrates (rice, bread pastry, cornbread, potatoes, and tubers such as yucca and ocumo) For the same reason the distribution of the carbohydrates of the diet was, in general, as follows 20 per cent at breakfast, 5 per cent at mid morning, 40 per cent at lunch, 10 per cent at 4 P M, and 25 per cent at dinner In those patients in which it was advisable not to omit insulin completely, only the regular type of this hormone was kept in use, administered to the patients 2 to 3 times a day according to the amount of glycosuria present before breakfast, lunch, or dinner (or both the latter) Regarding chlorpropamide, in most cases we used rather small doses, trying to determine the smallest

* The study reported here was supported in part by a grant from the Pfizer Corporation of Venezuela supplier of the chlorpropamide used

active one necessary to control the patients receiving it (TABLE 1) In a few cases with severe diabetes we prescribed larger doses but in no instance was any dose more than 500 mg 3 times a day We do not know whether the number of patients showing satisfactory response would have been larger if we had used larger doses or if we had given the drug for a longer period of time or both It is to be noted that in 3 cases it seemed that a better action of the drug upon the blood sugar was obtained if chlorpropamide was given 1 hour before meals It should be noted at this point that preliminary investigation in fasting diabetics showed that the action of the drug was more consistent 3 to 4 hours after it had been administered Owing to the short time available to complete this work it was decided to stop treatment after

TABLE 1
SCHEDULE OF TREATMENT WITH CHLORPROPAMIDE

Day of treatment	Milligrams		
	Breakfast	Lunch	Dinner
First	250	250	250
Second	250	125	250
Third	250	125	125
Fourth and beyond	125	125	125

8 to 10 days in cases with poor response We feel now that those patients who respond satisfactorily will do so after 5 to 7 days of treatment

Analysis of Results

The patients were grouped arbitrarily according to their responses as follows (TABLE 2)

Group A is composed of patients with excellent responses (no insulin needed) and includes those in good clinical condition who showed no glycosuria and had blood sugar values between normal limits (fasting and after meals)

Group B includes patients with good responses (no insulin needed) in whom fasting blood sugar is not more than 150 mg per cent and not more than 200 mg per cent 2 hours after each meal Slight glycosuria will exist but no acetonuria

Group C is made up of those showing a fair response (with or without insulin) and a fasting blood sugar between 150 and 200 mg per cent Blood sugar after meals is between 200 and 250 mg per cent with moderate glycosuria

Group D includes patients showing a poor response (with or without insulin) and a fasting blood sugar of more than 200 mg per cent and more than 250 mg per cent after meals Permanent and accentuated glycosuria is present with or without acetone in the urine

TABLE 2
VENOUS BLOOD SUGAR IN DIABETICS TREATED WITH CHLORPROPAMIDE

Type of response	Group average, mg per cent*			
	Fasting	2 hours after breakfast	2 hours after lunch	2 hours after dinner
Excellent (A)	115	118	136	125
Good (B)	140	156	189	185
Fair (C)	163	190	240	238
Poor (D)	225	260	320	296

* Figures were obtained by blood sampling each patient at least twice at each time but on different days

TABLE 3
RESULTS OBTAINED IN 52 DIABETIC PATIENTS TREATED WITH CHLORPROPAMIDE*

Response	No of cases	Percentages
Excellent (A)	10	19 2
Good (B)	18	34 6
Fair (C)	4	7 6
Poor (D)	20	38 4
Satisfactory results (group A plus group B)	28	53 8

* Age at onset of diabetes all ages

TABLE 4
RESULTS OBTAINED IN 30 DIABETIC PATIENTS TREATED WITH CHLORPROPAMIDE*

Response	No of cases	Percentages
Excellent (A)	9	30
Good (B)	16	53 3
Fair (C)	2	6 6
Poor (D)	3	10
Satisfactory results (group A plus group B)	25	83 3

* Age at onset 40 years or more

According to this classification (TABLE 3) we obtained satisfactory results in 53 8 per cent of the cases (groups A and B), and unsatisfactory responses in 46 0 per cent of the series (groups C and D) However, it is noteworthy that, in the group with poor response (20 cases, or 88 per cent of the group in which no satisfactory results were obtained), there were 5 patients who had developed diabetes before 10 years of age, 6 in whom the diabetes had had an onset between 11 and 20 years of age, and 6 in whom the onset age

TABLE 5
RESULTS OBTAINED IN 22 DIABETIC PATIENTS TREATED WITH CHLORPROPAMIDE*

Response	No of cases	Percentages
Excellent (A)	1	4.5
Good (B)	2	9.0
Fair (C)	2	9.0
Poor (D)	17	77.2
Satisfactory results (group A plus group B)	3	13.5

* Age at onset less than 40 years

TABLE 6
DATA ON 28 DIABETIC PATIENTS SHOWING EXCELLENT* AND GOOD† RESPONSE TO CHLORPROPAMIDE

Case No	Age at onset of diabetes (years)	Duration of diabetes (years)	Age (years)	Insulin U 24 hours	Body type (constitution)
1	30	3	33	0	Athletic
2	53	6	59	0	Asthenic
3	45	4	49	0	Pyknic
4	45	9	54	0	Pyknic
5	46	4	50	10	Pyknic
6	50	3	53	0	Pyknic
7	40	8	48	0	Pyknic
8	50	13	63	16	Pyknic
9	57	5	62	0	Pyknic
10	43	8	51	14	Pyknic
11	32	8	40	0	Pyknic
12	50	5	55	16	Asthenic
13	46	14	60	16	Pyknic
14	44	11	55	0	Asthenic
15	43	15	58	12	Asthenic athletic
16	50	18	68	26	Pyknic
17	49	5	54	12	Pyknic
18	44	2	46	12	Asthenic
19	42	3	45	10	Asthenic
20	57	2	59	0	Asthenic
21	42	7	49	16	Pyknic
22	51	12	63	16	Pyknic
23	41	12	53	14	Pyknic
24	37	10	47	10	Asthenic
25	51	17	68	20	Pyknic
26	48	17	65	16	Pyknic
27	40	6	46	18	Pyknic
28	44	8	52	16	Pyknic

* Cases 1 to 10
† Cases 11 to 28

had been between 31 and 40 years of age. As is known from the experience gained with other orally active hypoglycemic agents that have been studied failure of treatment is practically the rule in cases of diabetes of infantile, juvenile, and young adult types, this also seems to be the case with chlorpropamide. For example, if we take into consideration only those patients who had developed diabetes at 40 or more years of age (TABLE 4), we obtain

TABLE 7
DATA ON 24 DIABETIC PATIENTS SHOWING FAIR* AND POOR† RESPONSE TO CHLORPROPAMIDE

Case No	Age at onset of diabetes (years)	Duration of diabetes (years)	Age (years)	Insulin U /24 hours	Body type (constitution)
1	7	3	10	40	Asthenic
2	45	1	46	10	Asthenic
3	28	3	31	30	Asthenic
4	49	1	50	0	Asthenic
5	50	7	57	24	Asthenic
6	14	2	16	36	Pyknic
7	4	1	5	12	Pyknic
8	8	6	14	54	Asthenic
9	17	12	29	52	Asthenic
10	26	8	34	54	Asthenic
11	25	10	35	50	Asthenic
12	7	5	12	46	Asthenic
13	50	4	54	8	Pyknic
14	15	17	32	52	Asthenic
15	7	6	13	58	Asthenic
16	30	6	36	30	Asthenic
17	25	4	29	46	Athletic/asthenic
18	48	13	61	64	Pyknic
19	16	4	20	64	Athletic
20	16	9	25	48	Asthenic
21	27	6	33	30	Asthenic/athletic
22	15	8	23	60	Pyknic athletic
23	9	3	12	42	Asthenic
24	30	1	31	40	Asthenic

* Cases 1 to 4
† Cases 5 to 24

83.3 per cent of satisfactory results, this percentage is only 13.5 in patients with diabetes developing before 40 years of age (TABLE 5).

Examination of TABLE 6, which shows data on patients satisfactorily controlled (excellent and good results) with chlorpropamide, reveals that (1) nineteen (68.2 per cent) showed a body constitution of the pyknic type, (2) twenty-five (89.2 per cent) developed diabetes at 40 or more years of age, and (3) except for 1 patient, none received more than 20 U. of insulin every 24 hours.

On the other hand, TABLE 7, showing data on patients in whom unsatisfactory results were obtained, reveals that (1) only 4 patients (16 6 per cent) had a body constitution of the pyknic type, (2) in 19 cases (79 1 per cent), the diabetes had developed before 40 years of age, and (3) only 4 patients (16 6 per cent) needed less than 20 U of insulin per day for control

Tolerance and Side Effects

Generally speaking, the drug was well tolerated at the doses used One patient complained of epigastric distress, moderate pruritus without any apparent lesions was present in another In a girl 10 years of age there was a slight decrease in the white blood cell count, but without concomitant neutropenia In one instance, few red cells were found in the urinary sediment A male patient 23 years of age, with severe diabetes, complained of somnolence, pruritus of the eyes, and moderate headache However, we really do not know whether these symptoms were due to failure to control the diabetes during the observation period or to the use of the drug Treatment of a young woman 31 years of age, to whom we gave chlorpropamide at a dosage of 500 mg 3 times a day, was discontinued after 5 days because of epigastric pain Approximately 72 hours after discontinuance the patient had a rash all over the body, together with severe itching and slight fever (100° F) Antihistaminic drugs with small doses of prednisone improved the condition However, 24 hours later, there was an exacerbation of the skin lesions At this time a closer interrogation of the patient revealed that, as self prescribed treatment for a slight sore throat, she had taken penicillin tablets during the fourth and fifth day of the chlorpropamide treatment Allergist Marcel Granier was of the opinion that at least this second rash could be explained by sensitization to penicillin With proper treatment the patient recovered in a few days

Summary

A total of 52 nonselected patients were studied from 5 to 36 days Excellent results were obtained in 10 cases (19 2 per cent), good results in 18 patients (34 6 per cent), and fair results in 4 cases (7 6 per cent) Results obtained were poor in 20 cases (38 4 per cent), however, it should be noted that of these 20 cases, 17 were patients under 40 years of age, 11 of whom had developed diabetes when they were less than 20, the other 6 had developed it in their 20s As is well known, it is precisely in this type of diabetic that the oral hypoglycemic drugs have been less effective It seems that, in a high percentage of diabetics who respond well to chlorpropamide, it is possible to maintain control with a dosage much smaller than that required for similar drugs Many of the patients studied could be kept under control with only 125 mg administered after breakfast, lunch, and dinner—a total dosage of 375 mg During treatment, 1 of the patients showed a few red blood cells in the urinary sediment, another complained of a slight pruritus without an apparent lesion, and Case 12 showed somnolence, ocular pruritus, and a slight headache It is possible that these untoward effects were caused by a diabetic unbalance rather than by the direct action of the drug One case

showed an intense dermatitis about 72 hours after having had to discontinue chlorpropamide because of epigastric distress nevertheless it should be noted that this patient ingested 3 tablets daily of oral penicillin during the fourth and fifth days of treatment with chlorpropamide

Acknowledgments

We thank Rafael Nicanor Silva and Nidia M Fernandez for their help in the preparation of the manuscript and Ramon Cuevas for his technical assistance

CONSIDERATIONS FOR CONTROL NORMS TO EVALUATE RESULTS OF TREATMENT IN DIABETES MELLITUS

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If a real picture of the diabetic problem is to be obtained it must be considered not only in terms of the present and in those of the individual but also because of the increasing incidence of this disease its longer duration and its hereditary characteristics in terms of the future and of mankind

Before 1921 the average life expectancy of a diabetic was only 4 5 years after the disease had been discovered Today thanks to the discovery of the hormone insulin by F G Banting and C H Best and to many advances in different fields of medicine it is possible to say that if a diabetic is well controlled his life expectancy will generally be the same as that of a non diabetic of the same age and sex and that his illness should not be an obstacle to enjoyment of a useful life However the question emerges When can we say that a diabetic is well controlled? We do not intend to discuss this point at length however it is frequently said that if a diabetic has no sugar in the urine or shows only traces of glycosuria and if his blood sugar remains near normal ranges he can be considered well controlled This could be true in many cases but we frequently see patients whose diabetes could be considered well controlled according to these standards yet who suffer from different degenerative conditions that we think are of diabetic origin

In other words we think that hyperglycemia or glycosuria or both are indexes of the diabetes but cannot be taken as measures of the severity of the disease In fact we often see patients with diabetes of long duration who have had no proper medical care for years who show high values of sugar in the blood and in the urine and still are free from the so called diabetic complications At the same time unfortunately we see patients who adhere faithfully to the doctor s instructions show no glycosuria have moderate blood sugar levels most of the time but in whom degenerative conditions of the kidney and retinal vessels can be detected What is more important in many of these cases is that the diabetes or at least the hyperglycemia has not been present for a long time We say this because we feel that diabetes represents a very complex metabolic disturbance of which glycosuria or hyperglycemia or both are manifestations—the most frequent perhaps but not always the most important

With the discovery of orally active hypoglycemic agents it seems to us the problem has become even more complex

One thing we already know in many diabetics blood sugar levels can be maintained between satisfactory ranges with the use of the above mentioned drugs

However another question arises Can these patients be considered well controlled simply because their blood sugar is normal or almost so? It is

our belief that we must wait five to ten years before we can describe the condition of these patients accurately

The situation that confronts physicians and patients in 1958 is quite different from that when insulin was first made available insulin is a biological product and at that time it was unique as a diabetic specific In other words from now on the results obtained with any drug in cases of diabetes mellitus should be analyzed from the following points of view Does it satisfactorily lower the blood sugar? Does it have a better value than insulin in the prevention of the degenerative conditions? Does it have any undesirable effects? Can it be given safely for years to diabetics who often have fragile kidneys and liver?

Briefly we feel that in order to obtain better data to judge the results obtained in the treatment of diabetes it will be worthwhile to determine the type of control (such as laboratory tests X ray films and electrocardiograms) that should be used and the frequency with which it should be employed This would enable us to detect diabetic complications as early as these medical techniques are capable of uncovering them it would be the only way to determine when to readjust our therapeutic approach without the need of waiting for the appearance of full fledged clinical pictures such as those we see in cases of obstructive arteriopathy intercapillary glomerulosclerosis and advanced neuropathy It is our feeling that we may not have been quite correct in our treatment of some of our patients simply because our main criterion has been that of attaining satisfactory blood sugar levels

The problem is so complex and of such great significance that possibly it would justify the establishment of an international committee to coordinate the work of researchers in this field in different countries

Summary

In view of the current nature of the problem of diabetes and its treatment the evolution of this condition and the complications attendant on it (particularly those of a degenerative type), I consider it important to establish general norms for such aspects as medical controls and collateral studies These would permit not only judgment of the effectiveness of the treatment with reference to hyperglycemia or glycosuria or both but would also provide an opportunity to gather data on such factors as incidence degree of seriousness and evolution of the so-called complications of diabetes in relation to the various types of treatment At the same time in order to evaluate the action of new drugs administrable *per os* and capable of lowering the glucose levels (but not necessarily controlling other diabetic manifestations) it will be necessary to plan special studies to determine their control of hyperglycemia or glycosuria and also comparative studies of patients treated for years with the new drugs and of those patients treated with insulin It will also be necessary to establish the type and frequency of tests for the purpose of eliminating collateral or untoward reactions (or those of any other nature) that the hypoglycemic drugs active by the oral route are capable of producing

THE CLINICAL RESULTS OF THE TREATMENT OF DIABETES MELLITUS WITH CHLORPROPAMIDE

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During the last few years numerous studies have been made to find an

However it is evident that the relatively low toxicity of the compounds especially of tolbutamide as well as their usefulness for a specific group of diabetics can be taken for granted In a recent report [1] we summarized the results obtained with carbutamide and tolbutamide in the treatment of a group of 26 diabetics over a period of 21 months in general the results confirmed those of other investigators

Only 29 of the 35 patients have been treated for 4 months or more we therefore base our conclusions on these cases only Although we think that the treatment of the remaining patients can be considered a success data on them will not be included in this report as proof or justification of the conclusions

Of the 29 diabetics treated 14 are female and 15 male 35 to 68 years old with an average age of 55 1 years

6 months before preparation of this paper) and 2 had diabetes of 18 years duration

In the males 35 to 68 years old with an average age of 52 8 years the average duration of the disease was equally advanced (6 1 years) with one case of 23 years and 4 of less than 6 months (FIGURE 1)

Of the 29 diabetics 14 had been treated with tolbutamide for 12 months or longer at doses between 500 and 2000 mg daily 10 used between 10 and 80 U of insulin daily (average 43 U) and only the 5 remaining cases were maintained on diet alone

In all cases the treatment with the new drug began abruptly after preliminary clinical examination and analytical tests Except for Case 9 on

two occasions, the treatment was ambulatory and without change in the daily routine or in the diet previously followed

Among the patients previously treated with insulin were two (cases 4 and 6) who were positively refractory to tolbutamide as we found in an earlier trial

Regarding the others, one female patient with a serious diabetic retinopathy began the treatment in a state of acidosis another had an acute anthrax infection another had a carcinoma of the upper lip that was operated after the first week of treatment and two had tuberculosis of the lung (one cavitary)

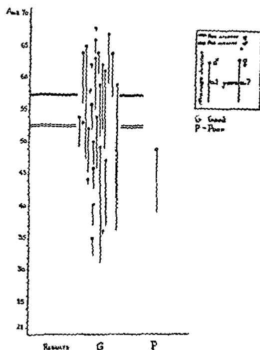


FIGURE 1 Relation of age of patient and duration of diabetes to response to treatment.

One case of cortico-adrenal hyperplasia with metabolic hyperfunction of Cushing's type and another with a pituitary chromophobe adenoma should be mentioned One patient developed dry gangrene of one foot during the sixth month of treatment but, in spite of this severe complication, we did not stop the medication Thus far he has had good results with regard to blood glucose levels

As coexistent illnesses, which may be considered minor as far as their relation to diabetes is concerned, essential hypertension was present in four patients and myocardial fibrosis in one

With the exception of two patients, all belonged to the pyknic or pyknic athletic groups

In addition to a complete clinical examination, the following analytical tests were made before the beginning of the trial

Glucose tolerance curve Using an oral dose of 25 gm glucose, analyses of blood glucose levels were made at 0-, 30-, 60-, and 120 min intervals The curve was repeated 48 hours later with an oral dose of 1 75 gm glucose per kilogram of body weight The blood glucose determinations were made by the Folin Kingsley Reynbold method until July and since then, by the Kingsley Reynhold Fister method

NPN determination This was determined by the hypobromite method

Hanger's Takata Ara's and Maclagan's tests In the majority of the patients and since June these tests have been performed by paper electrophoretic partition of serum proteins

Blood tests Hemoglobin determination hematocrit red and white cell counts and platelet count were carried out

After beginning treatment repeated blood glucose determinations were made in order to maintain close watch over the effect of the medication The other examinations were also repeated more or less frequently (an average of twice per month) in order to detect any possible systemic repercussions which might have developed On the basis of the described technique we think that the conclusions reached can be accepted with reasonable confidence

In the aforementioned experiments we endeavored to use the smallest initial dose possible Although we administered the drug in divided doses until the middle of February since then we have administered one dose daily at breakfast time

The maximum daily dose was 2 gm (4 tablets), and was used by only 3 patient the minimum dose in the initial treatment of 4 patients was 250 mg In 9 cases we gave 500 mg the average initial dose was 922 mg With high doses we observed no toxic reactions except dizziness and vertigo in 4 cases these reactions were coincident with hypoglycemia

In general these doses—particularly those of 1 gm or more—were administered for short periods usually not longer than 3 days The single exception was Case 2 a patient with acidosis to whom we gave this dosage for 3 months

The maintenance dosage usually reached at the end of the first week did not exceed 500 mg (4 cases) 12 patients were perfectly controlled on 125 mg daily and for 3 a daily dose of 62 mg was sufficient The average maintenance dosage was 248 mg

In regard to food all patients followed a liberal diet of carbohydrates with quantities ranging between 300 and 400 gm per day The other elements were relatively balanced We took special care to recommend to the patients that they should not vary the qualitative or quantitative substance of their diet from what they had been following before the beginning of these studies

In view of this we think that we can present the following conclusions with reasonable assurance However because of the short time and the limited number of patients the results should be considered as preliminary

Conclusions

(1) Chlorpropamide proved active in patients who previously benefited from other types of sulfonylureas (carbutamide and tolbutamide)

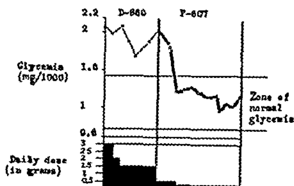


FIGURE 2

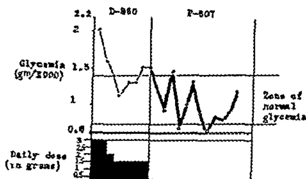


FIGURE 3

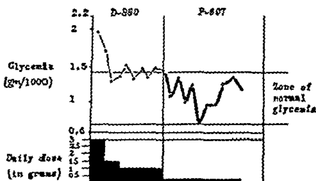


FIGURE 4

(2) Chlorpropamide was effective in two cases which had previously proved refractory to carbutamide and tolbutamide

(3) The clinical and metabolic regulation of diabetes with chlorpropamide is more simple and more complete than with other drugs tested so far (FIGURES 2 3, and 4) In the present group of patients, some noteworthy differ

ences have been verified regarding the interrelation of the results obtained and the ages of the patients and especially the time of evolution of the disease 37 per cent of the patients with good or excellent results are between the age of 61 and 68 (11 cases) with an average of 64 Greater differences occur in the relation with the duration of diabetes with 41 per cent of the patients having had the disease for 8 years or longer giving an average duration of 11 7 year (FIGURES 1 and 5) In our previous work with tolbutamide there were no good or fair results in the patients with diabetes of more than 8 years duration

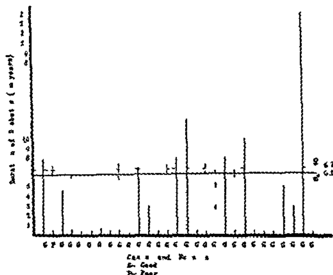


FIGURE 5 Duration of diabetes and response to treatment

(4) In addition to six cases where the drug showed no effect and that were not included in the present report it was found that chlorpropamide did not control the diabetes of a patient 49 years old whose disease had begun at 39 and whose treatment started when he was in a state of marked acidosis

This patient the only thin diabetic of the group had labile diabetes requiring a daily minimum of 60 U of insulin In spite of the fact that the patient had severe diabetic retinopathy we nevertheless maintained the treatment for 3½ months with doses varying between 1500 and 500 mg rapid disappearance of ketonuria and the establishment of blood glucose at lower (although still abnormally high) levels than at the beginning were verified From a systemic point of view there were no alterations worth recording except perhaps an improvement in visual acuity normalization of the serum proteins and small changes in the leukocyte count

(5) The coexistence of a severe infection anthrax or of latent or active tuberculosis (in the latter case bilateral and cavitary) does not adversely affect the efficient and rapid therapeutic action of chlorpropamide although

it should be noted that the doses used were slightly higher than those used for the other patients in general In one patient who dev... ...rene of one foot the drug nevertheless cont...

(6) On the average ... ain ing ... using ca

... objective tolerance we must report that fou... ...ed dizziness or vertigo coincident with hypoglycemia

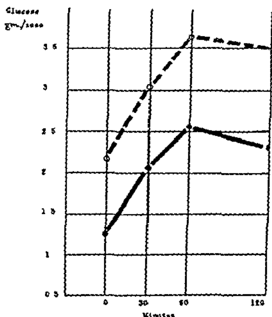


Figure 6 Glucose tolerance curve Broken line represents tolerance before therapy with chlorpropamide solid line after three months therapy with chlorpropamide Curves are averages of three cases previously treated with diet alone

In one case a diabetic doctor had a severe melancholic depression, with suicidal ideas that promptly disappeared after the carbohydrate content of the diet was increased No other neurological digestive or dermatological changes were observed in any of the patients of this group In one case not included in the present study the administration of chlorpropamide caused the appearance of a nettle rash in a patient who previously had shown sensitivity to tolbutamide and the sulfa drugs in general

Seven patients reported a sudden cephalic congestion after meals, with extrasystoles in two cases and mild lung edema in one—symptoms that disappeared promptly after stopping the intake of alcohol One of those who later resumed drinking wine did not have the same symptom We think the phenomenon may be due to a selective inhibition of acetaldehydic dehydrogenase

(8) Analytical observations showed that the blood glucose level remained generally normalized within the first week of treatment In some cases, where the patient spontaneously or upon our order did not adhere to the

the previous blood glucose level

An average of two urea determinations per month was made, and it was found that chlorpropamide did not increase the amount of nonprotein nitro

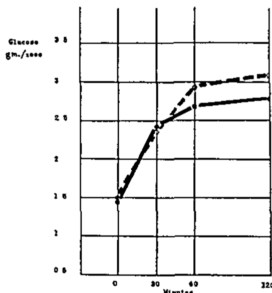


FIGURE 7 Glucose tolerance curve Broken line represents tolerance before therapy with chlorpropam de sol d l ne after three months theraj y w th chlorpropamide Curves are averages of four cases previously treated w th insul n

gen (NPN) On the contrary the NPN normalized itself or remained unchanged if previously high it was found too that a value as high as 76 does not per se mean a contraindication

Takata Ara s Hanger s and Maclagan s tests did not show undesirable

•

to be responsible for this abnormal finding

Serum protein electrophore is was performed in twenty one cases with normal results in all but one (Case 3)

No harmful repercussions were observe l in the l lood picture which was generally tested with the same frequency as blood urea On the contrary we saw the normalization of the blood picture in the cases that previously had

shown alterations (6 cases) It is worth mentioning that 1 patient, who started treatment with a severe leukopenia, showed a progressive increase of the white cells from 2300 to 4900 during the trial The hematocrit value decreased in 6 patients

If glycosuria was present this disappeared with the normalization of the blood sugar The same applies to albumin in two cases and to ketone bodies in two others In one patient traces of albumin persisted No pathological

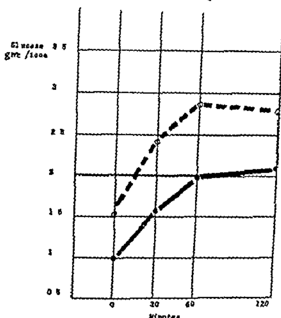


FIGURE 8 Glucose tolerance curve Broken line represents tolerance before therapy with chlorpropamide, solid line after three months therapy with chlorpropamide Curves are averages of seven cases previously treated with tolbutamide

changes in the sediment, especially crystalluria, and no alterations in the *p*H appeared

Glucose tolerance was investigated by means of an oral dose of glucose (1 75 gm /kg) and it was found that the 2 curves, obtained at intervals of 30 days or longer, can be considered (in their conjuncture) parallel (FIGURES 6, 7, 8, and 9) This shows that there is no modification in carbohydrate tolerance

It is our opinion that this new drug represents a major step in the treatment of diabetes mellitus [2] We know that more time, more patients, and above all more experience—clinical as well as analytical—are necessary in order to obtain conclusive knowledge about this therapy However, keeping in mind these limitations in comparison with the other drugs with which we have clinical experience (tolbutamide and carbutamide), we can say that chlor propamide is more active and less toxic in the therapeutic dosage we have used

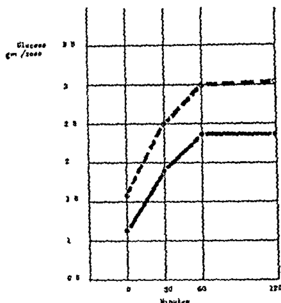


FIGURE 9 Glucose tolerance curve Broken line represents tolerance *before* therapy with chlorpropamide solid line after three months therapy with chlorpropamide Curves are averages of fourteen cases.

It is worth mentioning that all the patients were maintained on almost a free diet During this clinical trial we did not observe significant variations in the maintenance dosage

Acknowledgment

The authors wish to thank Ernesto Morais who permitted his laboratory to be used for the analyses mentioned in this report We also thank Pfizer International, Inc., New York N Y, M Usano of that firm, and Neo Farmacêutica Lda Lisbon Portugal for having placed at our disposal the quantities of chlorpropamide necessary for accomplishment of the study

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TOLERANCE FOR ALCOHOL IN PATIENTS ON CHLORPROPAMIDE

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By July 1, 1958 we had evaluated chlorpropamide clinically in 66 patients and had studied various sulfonylurea derivatives in approximately 600 patients with diabetes mellitus By August 30, we had increased the number to 624 patients who had been treated for more than 1 month, of these, 88 had received chlorpropamide Evaluating our results in this larger patient group, we can confirm the following five points relating to the control of diabetes mellitus with chlorpropamide

(1) The hypoglycemic activity of chlorpropamide, particularly with maximum therapeutic dosages, is in some instances superior to that achieved with carbutamide or tolbutamide

(2) The therapeutically effective dosage of chlorpropamide ranges from a maximum of 2 gm, which we used in some patients as an initial dose, to a minimum daily maintenance dose of 0 125 gm per day, these dosages were established on the basis of satisfactory daily control of blood sugar levels and the disappearance of sugar from the urine

(3) We observed a favorable response in 72 of 88 patients (80 per cent) and an inadequate response in 16 patients (20 per cent) It is important to note that 15 patients were less than 40 years old at the time of initiation of therapy, and that 12 of these were benefited by chlorpropamide therapy Only three showed an inadequate response

(4) With the initiation of therapy, and at intervals thereafter, patients were given various laboratory tests including liver function tests, blood counts, measurement of azotemia cholesterolemia, serum proteins, and serum transaminase From the results obtained by these laboratory tests, we observed no evidence that therapeutically effective dosages of chlorpropamide exhibited any type or degree of toxic effects

(5) Two types of side effects necessitated the discontinuance of therapy in some patients, all of whom belonged to the group that had not shown an adequate response to chlorpropamide The side effects were manifested as episodes of sensitivity and individual intolerance similar to those observed in response to other sulfonylurea derivatives and as episodes of specific intolerance affecting the gastrointestinal tract (nausea, sensation of abdominal fullness, constipation)

Initially, our attention was drawn to the unusual side effect associated with the ingestion of alcohol during chlorpropamide therapy by the fact that eight of twenty three patients who were under treatment complained that after meals they experienced one or more of a number of disagreeable symptoms that consisted of sensations of warmth, flushing, nausea, giddiness and, in some, tachycardia The patients had already come to the conclusion that

these aberrant symptoms were probably caused by the chlorpropamide, which had recently been prescribed for their diabetic condition, since the unusual sensations appeared about three or four days following initiation of therapy

The fact that these symptoms did not appear in all patients who were receiving chlorpropamide led us to the conclusion that this side effect was not due to the therapeutic action of the drug and therefore could not be considered a side effect that could be expected to occur as a direct consequence of its administration We reasoned, therefore, that it must be due to some set of circumstances peculiar to the patients who had developed the

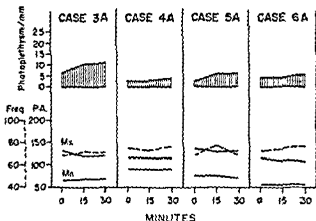


FIGURE 1 Alcohol test with normal subjects Symbols P A arterial blood pressure Freq heart rate (broken line) Mx maximum and Mn minimum

symptoms We began to investigate other factors and discovered that the phenomena appeared exclusively in those patients who were accustomed to drinking wine regularly during their meals We requested the patients to discontinue the ingestion of wine and their symptoms promptly disappeared When they resumed wine with their meals the symptoms returned

While we have had other sulfonylureas under study for some time as our total patient group shows, we have never before observed such a striking reaction to the ingestion of alcohol during treatment with a sulfonylurea It was necessary to discontinue chlorpropamide therapy in only one patient because of the development of this phenomenon Of course had this patient been satisfied to discontinue the ingestion of alcohol during chlorpropamide therapy, his symptoms would have disappeared and we should have been able to continue him under chlorpropamide treatment Such was not the case however and we substituted carbutamide for chlorpropamide three days after the initiation of chlorpropamide therapy On carbutamide, this patient experienced alcohol intolerance symptoms similar to those he had experienced while on chlorpropamide but they were mild in nature and disappeared after a few days on continued therapy Such a result corresponds to

the findings that some other investigators have reported concerning secondary effects observed in relation to the ingestion of alcohol by patients who were being treated with carbutamide or tolbutamide. Such symptoms of alcohol intolerance, however, have been infrequently observed with

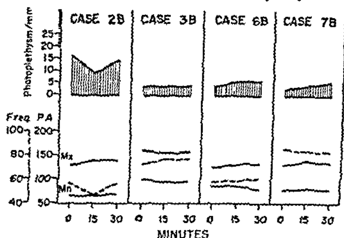


FIGURE 2. Alcohol test with diabetics not treated with chlorpropamide. Symbols as in FIGURE 1.

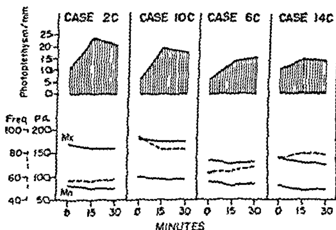


FIGURE 3. Alcohol test with diabetics under treatment with chlorpropamide. Symbols as in FIGURE 1.

these two drugs and are not considered to be of significance in their clinical application.

Following our observations of the eight patients who had experienced alcohol intolerance while under chlorpropamide therapy, we conducted the following clinical experiment.

Eight per cent (16 proof) ethyl alcohol was administered to all patients

in whom alcohol intolerance appeared These patients had taken chlorpropamide for at least 8 days and therapy which had successfully stabilized their diabetic state was being continued Within 10 to 20 min following the ingestion of the alcohol, these patients experienced an acute sensation of warmth of the face with flushing of the same area and in a few cases slight giddiness similar to that caused by ingestion of a large amount of alcohol

None of these symptoms was observed when the same dose of alcohol was given to normal subjects or to diabetics being treated with other sulfonylurea derivatives

This intolerance reaction to alcohol has been followed in all patients both before the ingestion
by measurement of
rate the auricular
graphic pattern

We have not observed any variation in the electrocardiographic pattern in any patient The values for systolic blood pressure have shown a slight tendency to reduce usually about 10 mm Hg while diastolic pressure tended to reduce approximately 5 mm Hg The pulse rates remained within normal limits and no significant variation from normal for each individual was observed The more relevant data recorded related to the behavior of the auricular photoplethysmographic wave FIGURES 1 2 and 3 show the changes that occurred in each of the patients studied

We can state that diabetics being treated with chlorpropamide show an intolerance to alcohol more frequently than the occasional occurrence that has been observed with other sulfonylureas However such side effects as do occur can be completely eliminated by discontinuing the ingestion of alcohol Thus the patient may make a choice between ingestion of alcohol or continuing therapy with chlorpropamide

THE TREATMENT OF DIABETES MELLITUS IN AREAS OF NUTRITIONAL DEFICIENCY

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The hypoglycemic effect of sulfonamides and derivatives of sulfonylureas has been observed by Janbon *et al* [1] since 1942, but it was not until 1955 that systematic application of such findings to the treatment of diabetes mellitus was started (in France) In the same year, systematic clinical trials were undertaken in Germany by Fuchs [2] Subsequently, the role of sulfonylurea derivatives in the management of diabetes mellitus has been extensively studied throughout the world [3 4] While the mode of action remains to be further clarified it is known that beneficial effects are obtained where the islets of Langerhans still possess some capacity to produce insulin [5 7] Hence, these drugs find particular application in the relatively mild maturity-onset type of diabetes [8]

Since most of the diabetics seen in the Philippines are of the maturity onset type the introduction of these oral hypoglycemic agents met with an enthusiastic response This was somewhat dampened by the realization that carbutamide (BZ 55) had a relatively high incidence of side reactions, while the better tolerated tolbutamide (D 860) was a less effective hypoglycemic agent

It is the purpose of this paper to describe our clinical studies on the new sulfonylurea derivative chlorpropamide (P 607)

Clinical Material and Method

The patients treated in the study described in this article were seen in the Diabetic Clinic of the Philippine General Hospital in Manila and in private practice They represent part of the group used in the investigation with carbutamide or tolbutamide or both A careful history was obtained and physical examination made in all instances Prior to therapy, base line determinations included fasting blood sugar levels urinalysis renal function studies hepatic function tests and hematological and adrenocortical evaluation Chest X rays (for pulmonary tuberculosis) and cardiovascular studies were also done The patients were seen in consultation by us at fortnightly intervals

There was no uniform plan of therapy Initial doses ranged from 500 to 2000 mg a day while maintenance doses ranged down to 125 mg daily The doses were adjusted according to the clinical and biochemical response of the patient

The 51 patients in the series were observed for periods of from 6 to 20 weeks at the time of the preparation of this report

Clinical Results

The response of a patient to therapy is difficult to gauge The following arbitrary designations were adopted The response was termed (1) excellent when the patient was rendered asymptomatic and where the fasting blood sugar level was lowered to and maintained within normal limits, (2) good

but where the fasting blood sugar, although not brought within 25 per cent

TABLE 1
RESULTS OF THERAPY WITH CHLORPROPAMIDE

Response	No of patients	Percentages	
Excellent	14	27 46	64 72
Good	19	37 26	
Fair	8	15 68	
Poor	6	11 76	19 60
Discontinued treatment	4	7 84	
Total	51	100 00	

of the normal levels could be maintained at least at 60 per cent of the initial levels and (4) poor when no clinical or biochemical improvement could be demonstrated In all instances side reactions were minimal These criteria were adopted because in almost all instances some degree of lowering of the blood sugar was observed, although it could not be maintained

TABLE 1 summarizes the results obtained with chlorpropamide in the 51 subjects studied 64 72 per cent showed excellent to good results 15 68 per cent had a fair response, 11 76 per cent were not benefited Four patients, or 7 84 per cent, were forced to suspend medication for various reasons

The following cases show the pattern of an excellent response Case 2 (FIGURE 1), F F 69 years female was recognized as a diabetic only 1 month before treatment She had the usual symptoms of polydipsia and polyphagia Initial fasting blood sugar was 248 mg per cent Treatment was started with 1500 mg of chlorpropamide divided into 3 doses There was prompt disappearance of glycosuria On the fourth day of treatment she complained of slight dizziness headache and blurring of vision Blood sugar was found to have dropped to 96 mg per cent, and treatment was suspended The disturbing symptoms promptly cleared Returning slight glycosuria and rising fasting blood sugar to 130 mg per cent indicated

toms subsided and blood sugar was maintained at 96 mg per cent After about 1 month, chlorpropamide was suspended to check the need for further maintenance There was a slight rise of FBS to 117 mg per cent in the next 3 weeks Resumption of the medication resulted in lowering of the blood sugar without reappearance of the hypoglycemic manifestations

Case 7 (FIGURE 2), A M, 54 years, male, was known to be diabetic for 2½ years and had required 30 to 40 U of insulin for the past 2 years An initial dose of 1000 mg chlorpropamide resulted in a precipitous drop of

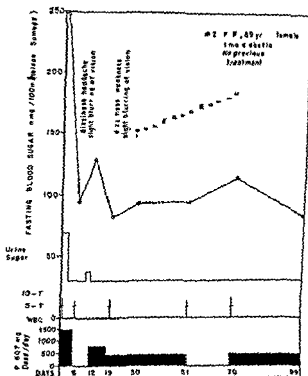


FIGURE 1

FBS from 210 mg per cent to 90 mg per cent in 2 days with accompanying dizziness slight weakness, and unsteadiness of gait These disappeared with suspension of treatment for about 5 days Reinstitution of the drug at a dosage of 500 mg daily produced a slow decline in the FBS from 105 to 69 mg per cent in 31 days, without reappearance of side effects Medication was suspended for 2 weeks and FBS rose to 85 mg per cent Resumption of treatment at 250 mg daily and, later, 178 mg daily maintained blood sugar between 75 mg and 90 mg per cent

FIGURE 3 shows an example of a good response Case 19, D T, 42 years, was found to have diabetes mellitus when he was confined for a pneumonitis Initial blood sugar was 249 mg per cent This was controlled with NPH

insulin at dosages reduced stepwise from 25 to a maintenance of 10 U daily At the end of 1 month, FBS was maintained at 62 mg per cent, and insulin was suspended After 1 week, FBS had risen to 125 mg per cent and carbutamide was given at levels of 500 mg daily This caused a drop of the FBS to 75 mg per cent in the next 4 weeks Carbutamide was suspended when FBS was reduced to 69 mg per cent The patient was next seen about 2½ months later when he observed returning glycosuria FBS had risen to 138 mg per cent, and carbutamide was resumed at 750 mg daily

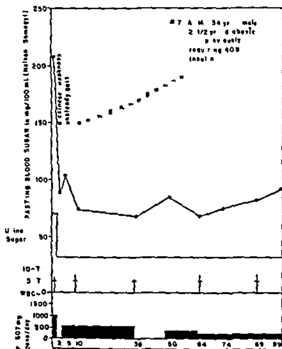


FIGURE 2

Blood st
previous
to it at
glycosuria the FBS showed a slow tendency to rise to 77, then 94 and, finally, 112 mg per cent The dose was then adjusted to 750 mg daily Blood sugar showed a prompt fall At one visit, a sudden elevation of FBS to 143 mg per cent was found to be associated with mental strain Use of a tranquilizer without further adjustment of diet or increase in chlorpropamide dose, caused a return to the former state of control

FIGURE 4 depicts the course of one of the youngest diabetics in our series Case 20 O J, was first diagnosed at the age of 23 years He is a male and

had pulmonary tuberculosis concomitantly. He did not receive any systematic form of treatment during the first 7 years of his diabetic life, and initial treatment consisted of 2000 mg. carbutamide daily. In 2 weeks FBS dropped from 260 mg. per cent to 143 mg. per cent, and glycosuria declined from heavy to moderate. Carbutamide was reduced to 1500 mg. daily because of the appearance of itching skin rashes. About 2½ months later, medication was suspended because of the reappearance of the cutaneous

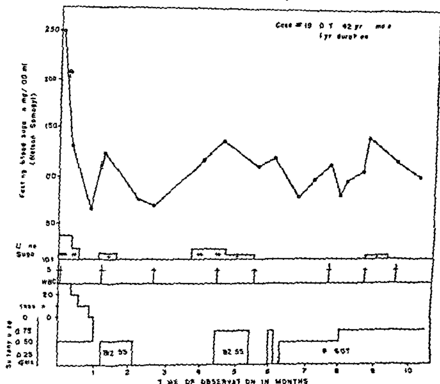


FIGURE 3

rash and a tendency to lowering of the leukocyte count. During this period, blood sugar remained about 150 mg. per cent. After 1 month on antihistamines the skin condition had cleared and the white blood cell count (WBC) was normal, carbutamide was resumed. Failure to lower the FBS below 150 mg. per cent, together with appearance of epigastric pain and diarrhea, caused suspension of carbutamide after 2 months. NPH insulin was given at levels of 15 U. daily. This, too, simply maintained FBS at the same level. After 2 more months the patient complained of the injections and requested another trial with oral hypoglycemics. Tolbutamide was then available and a trial was instituted at a dosage of 1500 mg. daily. Gastrointestinal disturbance, sore throat, and leukopenia ensued and caused immediate suspension of the drug; it was necessary to resume insulin. A

combination of 15 U NPH insulin and 1000 mg of carbutamide produced a progressive lowering of the blood sugar far more efficiently than could be produced by either drug alone

The insulin carbutamide combination was suspended when chlorpropamide became available Started at 500 mg daily chlorpropamide proved insufficient and the dose was gradually increased to 1000 mg daily Although the blood sugar could not be maintained at the same level as that obtainable with the insulin carbutamide combination the effects were definitely better

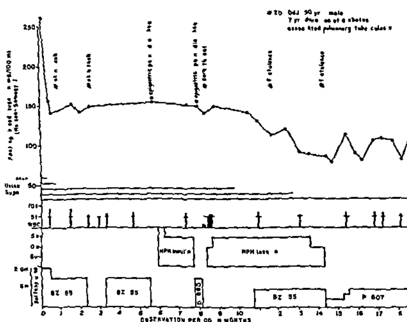


FIGURE 4

than with either 15 U of insulin or 1500 mg of carbutamide alone We plan to combine a small amount of insulin with chlorpropamide

FIGURE 5 shows an example of a poor response Case 33 V I is a 51 year old male who had diabetes for 9 years and required 40 U of insulin for control He showed evidence of hepatic dysfunction particularly in the abnormal results of flocculation tests He was started on 750 mg of chlorpropamide daily Because of lack of response dosage was increased to 1500 mg daily There was a slight drop of FBS from 225 mg per cent to 148 mg per cent but without changing the dose or diet, the blood sugar could not be maintained at this level The addition of 15 U of insulin arrested the upward spiraling of the blood sugar level but could not reduce it to normal levels

FIGURE 6 represents the interesting case of a 58 year old female who was

known to be diabetic for 2 years, and had been treated with diet and insulin She was admitted to the hospital for a lower abdominal mass, diagnosed by the Department of Gynecology as a cystadenoma of the ovary Surgical intervention was projected, and the patient was referred for control of the diabetic picture prior to surgery When first seen, she had a blood sugar level of 194 mg per cent, heavy glycosuria, and pruritus vulvae She was placed on 2000 mg of chlorpropamide daily FBS dropped to 112 mg per

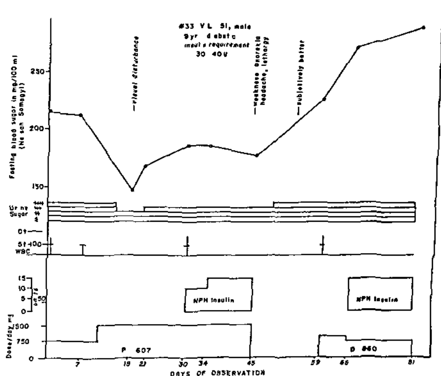


FIGURE 5

cent in 3 days and glycosuria was reduced to a minimum A panhysterectomy was performed the next day, and the postoperative course was uneventful The diabetic picture remained under control Two weeks later it was necessary to reduce the medication to 1000 mg daily because of a transient weakness and dizziness Attempts to reduce the dose further resulted in a rise of the blood sugar, a maintenance dose of between 750 mg and 1000 mg daily was found adequate

The pattern of response of the patients treated in our study was very encouraging There is a uniform tendency to lower the initial hyperglycemia and to maintain the lowered blood sugar level FIGURE 7 depicts the relationship between the blood sugar level in patients whose response was rated fair to excellent and the amount of chlorpropamide required to

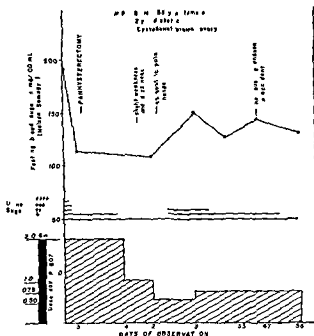


FIGURE 6

FIGURE 7

produce those results Initial doses of slightly more than 1.5 gm daily were sufficient to produce a reduction of the fasting blood sugar from an average of about 180 mg per cent to about 118 mg per cent in 1 week. The fasting blood sugar could be maintained at about 100 mg per cent by maintenance doses as low as about 400 mg daily. These doses are certainly very much smaller than those we utilized previously with the other sulfonyl urea derivatives.

Side Effects

The patients were carefully questioned at each visit about any untoward effects. TABLE 2 shows the side reactions that were encountered in the

TABLE 2
SIDE REACTIONS OBSERVED WITH CHLORPROPAMIDE THERAPY*

Reactions	No. of cases
Transient weakness	13†
Transient dizziness and headache	9
Transient blurring of vision	8
Unsteadiness of gait	3
Sialorrhea	3
Increased hunger pains	2
Flatulence	2
Localized dermatitis	1
Exfoliative dermatitis	1†
Generalized allergy	1†
Leukopenia	0
Purpura or anemia	0

* Duration of treatment of 51 patients 4 months.
† Suspension of chlorpropamide therapy caused disappearance of symptoms.

51 patients during the period of observation. The most frequent complaints were those of transient weakness, dizziness, headache and blurring of vision. They occurred with greatest frequency at the start of the investigation, when we used doses of chlorpropamide approximating those of tolbutamide. The complaints were often registered about 3 to 5 days after onset of treatment, and promptly cleared when medication was suspended. The effects have been attributed to a rapid reduction of the blood sugar levels as shown by close observations and repeated blood sugar checks similar to those depicted in FIGURES 1 and 2. Reduction of the initial doses of chlorpropamide to an average of 750 mg a day with slight adjustment of the dose according to the individual response of the patient, diminished the frequency of the complaints in subsequent patients.

Four patients did not continue treatment. In 2 patients the appearance

fourth developed exfoliative dermatitis during the course of treatment This is graphically shown in FIGURE 8 Case 36 R V 41 years female was known to be diabetic for 10 years and had an insulin requirement that varied from 20 to 40 U daily She also had pulmonary tuberculosis and a family history of allergic tendency although the actual allergens could not be pinpointed Initial FBS was 190 mg per cent and she was started on 750 mg chlorpropamide daily Weekly visits thereafter showed progressive

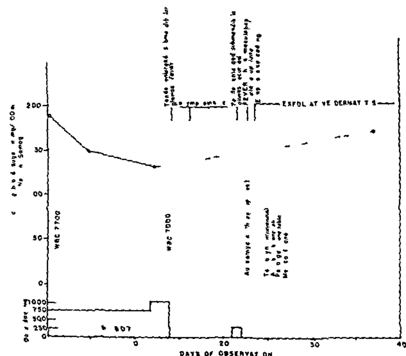


FIGURE 8

lowering of the blood sugar levels to 143 mg per cent and then to 130 mg per cent she was asymptomatic On her second visit the dose was increased to 1000 mg daily After 2 days she complained of slight fever and tender enlarged submandibular glands Blood examination showed normal WBC She was then advised to suspend chlorpropamide and all symptoms subsided 2 days later

After one week the patient again felt tenderness and enlargement of the submandibular glands and slight fever She took one tablet of chlorpropamide at noon and complained of heaviness of the body higher fever chills

continued to spread. Subsequent medication with Tetracyn, Antistine and Peralga brought no relief and her condition suddenly became a full blown exfoliative dermatitis. When seen in the hospital, she appeared in critical stage and had to be treated with insulin and Meticortelone. The events do not with certainty indict chlorpropamide as the culprit, but the case is being presented as one where a dangerous skin disease occurred during the course of treatment. The patient recovered completely from the exfoliative dermatitis.

TABLE 3
COMPARISON OF DIABETIC DEATH RATES OF DIFFERENT COUNTRIES

Country	Year	Population	Deaths from all causes	Deaths from diabetes	
				Number	Rate per 100 000
Australia	1949	7 912 638	75 260	1 473	18 6
United States	1950	150 697 000	1 450 270	25 010	16 6
Norway	1949	3 233 000	29 082	440	13 6
Canada	1950	13 821 000	123 649	1 545	11 2
France	1950	41 900 000	526 280	3 763	9 0
Italy	1950	46 279 500	448 971	3 544	7 7
Spain	1949	28 023 000	316 308	1 492	5 3
Mexico	1945	22 233 200	433 694	811	3 6
Colombia	1948	10 776 900	154 392	265	2 5
Japan	1950	83 200 000	908 801	2 007	2 4
Philippines	1956	21 856 875	219 719	137	0 6

TABLE 4
INCIDENCE OF DIABETES MELLITUS AND DIABETIC ACIDOSIS IN PATIENTS AT THE PHILIPPINE GENERAL HOSPITAL, MANILA, PHILIPPINES, 1945-1955

Number of hospital admissions	181 116
Number of medical cases	32 112
Admissions and cases diagnosed as diabetes mellitus	619 (1 9 per cent)
Admissions and cases diagnosed as diabetic acidosis	10

TABLE 5
COMPARISON OF INCIDENCE OF DIABETIC ACIDOSIS IN THE PHILIPPINES AND THE UNITED STATES

	Philippine General Hospital	New England Deaconess Hospital	Philadelphia General Hospital
Period covered	1945 1955	1946-1951	1931 1936
Number of diabetic admissions	639	10 200	3 009
Number of diabetic comas	10	153	268

TABLE 6
COMPARISON OF DIETARY INTAKE AND DIABETIC MORTALITY RATES IN DIFFERENT COUNTRIES

Country	Total calories	Distribution of caloric intake									Diabetes mellitus mortality per 100 000
		Cereal	Root tubers	Sugar	Fats	Legumes	Fruits and vegetables	Meat	Milk	Wine	
Australia	3128	872	93	582	420	28	106	755	282	—	18 6
United States	3249	887	139	515	502	105	210	524	367	—	16 6
Norway	3129	1147	242	391	631	27	40	326	313	12	13 6
Canada	3109	943	177	518	464	81	118	442	366	—	11 2
France	3012	1203	247	247	320	100	67	303	222	298	9 0
Italy	2627	1606	63	80	256	158	52	148	108	156	7 7
Spain	2788	1416	180	143	360	158	86	324	101	110	5 3
Mexico	1909	1004	42	197	106	108	96	197	155	4	2 6
Colombia	1934	768	313	98	93	45	100	279	233	5	2 5
Japan	2268	1359	157	163	41	147	83	107	11	—	2 4
Philippines	2021	1115	264	124	130	79	23	218	68	—	0 6

There were no evidences of impairment of renal function or liver function, or of injury of the bone marrow in any of the cases under observation

Discussion

Diabetes mellitus is not a major cause of death in the Philippines. TABLE 3 shows a comparison of pertinent figures available from different parts of the world[9] Among the countries on this side of the Pacific Australia ranks high with diabetes mellitus accounting for 18 6 deaths per 100,000 population Japan has only 2 4 per 100 000 population and the Philippines had only 137 deaths ascribed to the disease in 1956 or a mortality rate of 0 6 per 100 000 population

Figures from the Philippine General Hospital (TABLE 4) for the period 1945 to 1955 show that 1 9 per cent of the patients admitted to the Department of Medicine were diagnosed as cases of diabetes mellitus however of the 639 cases occurring during this period only 10 showed diabetic acidosis In comparison (TABLE 5) 5 year surveys in the New England Deaconess Hospital Boston Mass and the Philadelphia General Hospital Philadelphia Pa show a large number of patients with diabetic acidosis[9]

These statistics support the observation by most practitioners in the Philippines that the great majority of diabetics there are mild and that juvenile or growth onset type of diabetes is a rarity The factors that may explain this are not entirely clear One suggestion that may be tenable is that the Filipino diet[10] which is relatively low in fat and moderate in calories approximates the diabetic diet Inasmuch as dietary restriction is an accepted method of treatment it is postulated that the somewhat nutritionally deficient diet contributes to a milder course of the disease

TABLE 6 shows a comparison of the general food habits from different countries as gleaned from Food and Agriculture Organization (FAO) reports[11] There appears to be a rough correlation between the nutritional adequacy of the diet and the mortality rate for diabetes mellitus particularly with respect to the total calories and the fat content The evidence while not conclusive, points to a trend which deserves further investigation

The use of the sulfonylurea derivatives in diabetes mellitus ushers in a new era in management of the disease This is especially true in areas where the diabetes found is mild and where resistance and the incidence of coma are low Such conditions obtain in the Philippines and for this reason we have been very much interested in studying chlorpropamide

Summary

The great majority of cases of diabetes mellitus in the Philippines are of the maturity-onset type The incidence of diabetic acidosis and of juvenile diabetes is small

Chlorpropamide has shown great effectiveness in controlling diabetes mellitus

The dose of chlorpropamide found effective in controlling the disease is much smaller than expected initial dosages of 750 to 1000 mg daily and maintenance doses of 250 to 500 mg daily were found to be adequate

Few side effects were encountered The most frequent were symptoms of hypoglycemia produced when fairly large doses were employed

A case of exfoliative dermatitis was reported to have developed in one patient during the course of treatment

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THE PROBLEM OF DIABETES IN THE SINGAPORE POPULATION AND THE IMPACT OF ORAL ANTIDIABETICS ON ITS MANAGEMENT

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Singapore General Hospital, Singapore

In Singapore, where the population is made up of many races or ethnic groups and where literacy is far from universal, diabetes poses problems, particularly in management quite different from those met with in the more highly developed countries of America and Europe Singapore has a population of 1,500 000, consisting mainly of Chinese (76 4 per cent), Malays, and Indians It was from this population that subjects of this study were drawn and the investigation was carried out in 1 of the 2 teaching medical units in the Singapore General Hospital

The Size of the Problem

In 1957 the number of patients with diabetes newly registered in 1 of 2 diabetes clinics at the hospital totaled 377 These, together with patients already on the register gave a total clinic attendance of 7,217 for the year There is as yet no unified center for the treatment of diabetes, and such treatment is undertaken at the Singapore General Hospital the Tuberculosis Hospital, outpatient clinics of the outpatient service, and by private practitioners Therefore statistical data on the numerical size of the problem posed by diabetes mellitus are difficult to obtain However, there is little doubt that diabetes constitutes a large part of the problem of the care of the health of the population

Etiological Factors

While definitive etiological factors in cases seen in Singapore and elsewhere may be largely similar, the emphasis on any particular etiological factor seems to vary Thus, in Singapore, diabetes in children is decidedly uncommon, and the disease frequently manifests itself in middle life

The maximum incidence falls within the age group of 36 to 65 years The sex incidence also differs from that seen in other countries, as the disease is commoner among males than females In its distribution among the various races, the disease is commoner among Indians in a population predominantly Chinese The majority of patients comes from the low income group in which carbohydrates, especially in the form of rice, constitute the major part of the diet

Presenting Symptoms

Relatively few of our patients come to us because sugar was found in the urine on routine examination Thirst and frequent passage of large quantities of urine, especially at night comprise the symptoms most frequently complained of while not a few come for medical advice because they observed

ants gathering about their urine or the toilet Infective complications chiefly carbuncle and pulmonary tuberculosis cause them to be seen by our surgical colleagues or in the tuberculosis ho pital While a wide variety of complications is encountered the incidence of diabetic coma is definitely low In one of the two medical units at the General Hospital only seven patients in diabetic coma were seen in the past year This is not surprising as the great majority of the diabetics have diabetes of late onset

Problem in Management

It is generally known that the Chinese culture emphasizes good food and in Singapore this attitude towar l food is shared by the other communities Hence it is extremely difficult if not impossible to expect our patients to adhere to a strict diabetic diet for long It is chiefly for this reason that hospital in patient treatment with its dietary regimen is unsatisfactory, as the patient s home food differs both in quality and quantity from that given him in the hospital Consequently the disease is treated largely on an out patient basis where the patient is allowed a free diet Only those with infective and other severe complications are admitted to the hospital wards In outpatient treatment while sugar and sweetened foods are strictly pro hibited other carbohydrates (including rice) are offered at each meal In this way our patients are able to arrive at a fairly constant carbohydrate intake frequently higher than the intake they can be allowed on dietary treatment alone

As observed earlier the majority of our patients come from the poorer segment of the population and management of the disease and the initiation into a diabetic life are rendered more difficult by their illiteracy Printed instructions are of little value Each patient must be instructed in turn and where possible if language difficulties can be overcome some under standing of the disease process must be brought within his grasp If this is achieved the illiterate diabetic will adhere faithfully to the advice of the physician and his disease will be brought under control quickly Until the advent of oral drug therapy the overwhelming majority of patients received insul n treatment and were carefully taught to inject themselves and to test and chart their urinary sugar results

As soon as it became generally known that tablets were available for the treatment of diabetes almost every patient saw in this a relief from daily injections The demand for oral therapy was persistent and almost uni versal in the d abetes clinics and carbutamide and tolbutamide were tried and reported on by Tan Bock Yam and Ronald Wells (1957)

Clinical Trial with Chlorpropamide

In Singapore when chlorpropamide became available for invest gational use it was decided to conduct a clinical trial in an attempt to assess its efficacy in the treatment of diabetes mellitus

Material Ther

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However it was felt that patients with infective complications or acidosis or both should be excluded as the trial was conducted on an outpatient basis so that direct supervision could not be as close as when patients were hospitalized

The age of patients at the onset of diabetic symptoms and their sex and race distribution are shown in TABLES 1 and 2

TABLE 1
AGE AT ONSET OF DIABETES AND SEX DISTRIBUTION OF PATIENTS ON CHLORPROPAMIDE

Age*	26–30	31–35	36–40	41–45	46–50	51–55	56–60	61–65	66–70	Total
Male	2	6		7	6	8	4	1	1	42
Female	0	0	5	5	9	3	1	1	1	25
Total	2	6	12	12	15	11	5	2	2	67

* Average age at onset of diabetes 45 8 years

TABLE 2
RACE DISTRIBUTION OF PATIENTS ON CHLORPROPAMIDE

	No of patients*				
	Chinese	Indian	Malay	Others	Total
Group A	12 (44 4)	13 (48 1)	nil	2 (5)	27 (100)
Group B	11 (44 0)	9 (36 0)	4 (16 0)	1 (4 0)	25 (100)
Group C	2 13 3)	9 (60 0)	4 (26 7)	nil	15 (100)
Total	25 (37 3)	31 (46 3)	8 (11 9)	3 (4 5)	67 (100)

* Figures in parentheses are percentages of total

1 tho e [illegible]
were treated with tolbutamide and Group C those treated with insulin prior to chlorpropamide therapy

All patients were examined clinically and urinalyses glucose tolerance tests X ray examinations of the chest and blood counts were done the first and last were repeated at intervals

The patients attended the clinic weekly while dosage of chlorpropamide was adjusted according to the urinary sugar reaction to Benedict s reagent the urinary tests (checked by the laboratory at intervals) were done by the

TABLE 3

CLINICAL DATA OF 27 DIABETES PATIENTS (GROUP 1) NOT ON ANY PREVIOUS TREATMENT

Case No	Reference No	Race*	Sex	Present age	Age at onset (years)	Blood sugar mg % Fasting	Blood sugar mg % Max †	Final Chlorpropamide dosage gm/day	Duration of treatment (weeks)	Quality of control	Side effects	Thrombocytes per cu mm blood	Remarks
1	N1875	Ind	M	33	32½	138	206	0.5	22	good	Furuncles giddiness	180 000	
2	N1429	Ind	M	50	50	246	416	0.75	14	good	giddiness initially	—	
3	N2261	Ind	M	44	41	288	312	0.7	21	good	epigastric discomfort	130 000	
4	N2253	Ch	F	53	51	235	392	1.0	17	fair	nil	110 000	random blood sugar 152 mg %
5	N2467	Ind	M	55	53	306	340	0.5	15	good	nil	225 000	random blood sugar 61 mg %
6	N3140	Ch	M	54	49	216	294	0.5	12	good	sleepiness	135 000	
7	N3246	Ch	F	54	34	218	310	0.75	4	poor	nil	—	
8	N3247	Eur	F	54	49	216	312	0.5	6	good	constipation	130 000	
9	N3250	Ind	M	56	54	243	408	0.5	9	good	giddiness	140 000	cataract
10	N3 69	Ch	F	51	50	204	334	0.5	11	good	nil	80 000	
11	N3357	Filp	F	49	49	86	217	0.5	10	good	nil	200 000	
12	N2227	Ch	M	69	61	210	—	0.25	14	good	anorexia	80 000	hypertension
13	N3440	Ch	M	51	47		—	0.75	10	good	nil	80 000	
14	N3502	Ind	M	35		90	236	0.5	8	good	nil	165 000	palpitations
15	N3505	Ch	F	33	51	184	280	0.5	8	good	nil	100 000	
16	N3237	Ind	M	49	40	172	263	0.5	6	good	nil	—	
17	89 37	Ch	F	51	46	212	370	1.0	4	poor	profuse sweating	—	Pt refused to continue with P607
18	N3819	Ind	M	38	37	233	444	0.5	7	fair	nil	—	
19	N3972	Ind	M	39	30	234	370	0.75	5	good	constipation	—	
20	N4024	Ch	M	53	52	—		1.0	4	good	nil	—	furuncles
21	N3443	Ind	M	37	34	128	212	0.25	3	good	nil	—	
22	N4001	Ch	M	39	37½	227	392	1.0	4	good	nil	—	
23	N3664	Ind	M	44	42	332	412	0.5	8	good	constipation	250 000	peripheral neuritis
24	N1499	Ind	F	46	45	184	302	0.75	8	good	nil	139 000	
25	N4178	Ch	M	38	38	238	366	1.0	4	poor	nil	—	
26	N3620	Ind	M	38	38	—		0.5	6	good	nil	150 000	appendix abscess
27	N3361	Ch	F	40	39	—	—	0.5	8	fair	nil	60 000	cataract both eyes

* Key Ind Indian, Ch Chinese Eur European Filp Filipino
† In glucose tolerance test

TABLE 4
CLINICAL DATA OF 25 DIABETES PATIENTS (GROUP B) PREVIOUSLY TREATED WITH TOLBUTAMIDE

Case No	Reference No	Race*	Sex	Present age	Age at onset	Blood sugar mg %		Final dosage		Duration of treatment		Quality of control		Side effects		Thrombocytes per cu mm blood	Remarks
						Fasting	Max †	Chlorpropamide gm/day	Tolbutamide gm/day	Chlorpropamide (weeks)	Tolbutamide (months)	Chlorpropamide	Tolbutamide	Chlorpropamide	Tolbutamide		
28	1878/56	Ch	M	67	62	130	340	1.0	1.0	2	12	poor	good	weak, tired, anorexia	nil	—	patient refused to continue with chlorpropamide
29	N812	Malay	M	31	28	220	370	0.75	0.5	11	18	fair	fair	nil	nil	130 000	
30	2335/56	Ind	M	52	44	283	444	0.5	1.5	7	16	fair	fair	nil	nil	140 000	
31	N1413	Cey	M	52	47	233	396	0.25	1.5	12	9	good	fair	nil	nil	120 000	
32	N3145	Ch	F	37	37	106	243	0.75	1.0	6	1	fair	good	erythematous rash	nil	280 000	
33	462/57	Ind	M	54	51	256	555	0.75	1.5	4	2	poor	poor	nil	nil	—	unreliable
34	N833	Ch	F	52	50	—	—	1.0	1.5	9	8	good	good	purpuric rash	nil	180 000	
35	N1773	Ind	M	46	42	142	242	0.5	1.0	12	3	good	good	erythematous rash	nil	60,000	
36	1394/57	Ch	F	55	48	—	—	0.75	1.5	5	12	fair	poor	nil	nil	—	
37	N1187	Ind	M	65	60	200	—	0.5	1.5	11	6	good	fair	constipation	nil	—	
38	2387/56	Malay	M	37	35	200	284	0.75	1.5	12	15	better	good	nil	nil	200 000	
39	N1334	Ch	M	57	53	200	285	0.75	1.5	11	8½	good	fair	nil	nil	210 000	
40	651/57	Ch	F	47	46	—	—	0.5	1.5	13	11	good	good	giddy	nil	160 000	
41	N1567	Ch	F	74	69	120	240	0.5	1.5	2	4	poor	fair	marked giddiness	nil	—	chlorpropamide discontinued
42	N908	Ch	F	52	42	152	270	0.75	1.5	8	16	good	good	nil	nil	150 000	
43	N655	Ind	M	56	54	350	714	0.5	0.5	10	10	good	good	constipation	nil	90 000	random blood sugar 121 mg %
44	N597	Sikh	F	56	44	250	362	1.0	1.5	6	7	fair	good	nausea vomiting	nil	—	chlorpropamide discontinued
45	609/56	Ind	M	54	51	178	392	1.0	1.5	9	12	good	good	nil	nil	180 000	
46	N1652	Ch	F	30	49	356	540	1.0	1.5	7	10	good	fair	erythematous rash	nil	110 000	
47	705/56	Ch	F	44	41	200	316	0.5	1.5	9	13	good	good	nil	nil	160 000	
48	N595	Malay	F	47	45	—	—	0.25	0.5	7	6	good	good	nil	nil	130 000	random blood sugar 133 mg %
49	N837	Malay	F	38	35½	224	354	1.0	1.5	6	13	good	good	constipation	nil	—	
50	N503	Ind	F	65	62	217	392	0.75	1.5	13	10	good	fair	nil	nil	200 000	
51	1080/56	Ch	F	53	49	167	350	0.75	1.5	6	28	good	fair	nil	nil	—	
52	N615	Ind	M	35	31	—	—	0.75	1.5	5	10	good	fair	nil	nil	—	

* Key: Ch, Chinese; Ind, Indian; Cey, Ceylonese.
† In glucose tolerance test.

TABLE 5
CLINICAL DATA OF 15 DIABETES PATIENTS (GROUP C) PREVIOUSLY TREATED WITH INSULIN

Case No	Reference No	Race*	Sex	Present age	Age at onset	Blood sugar mg %		Final dosage		Duration of treatment		Quality of control		Chlorpropamide side effects	Insulin shock	Thrombocytes per cu mm of blood	Remarks
						Fasting	Max †	Chlorpropamide gm / day	Insulin U / day	Chlorpropamide (weeks)	Insulin (months)	Chlorpropamide	Insulin				
53	N702	Ind	M	43	42	160	362	1 5	SI 40 PZI 48	5	11	poor	good	giddiness weight loss anorexia lassitude	nil	—	chlorpropamide discontinued
54	N1034	Ch	M	64	60	—	—	0 75	SI 40 PZI 24	20	5½	good	good	constipation	coma once	110 000	retinopathy and cataract
55	N2123	Ind	M	39	33	280	362	0 25	IZS(L)28	38	2	good	poor	nil	nil	165 000	random blood sugar 93 mg %
56	N2386	Ind	M	51	50½	206	388	1 0	IZS(L)40	9	5	good	poor	giddiness initially	coma once	140 000	
57	N668	Ind	M	45	44	174	312	1 0	IZS(L)136	20	9	good	fair	nil	nil	160 000	insulin gradually withdrawn
58	N1873	Ch	M	39	39	146	—	1 0	IZS(L)24 IZS(SL)24	13	2	good	fair	nil	nil	150 000	random blood sugar 111 mg %
59	2134/57	Malay	M	48	45½	166	333	0 75	IZS(L)44	9	2½	good	fair	nil	nil	150 000	random blood sugar 86 mg %
60	N1724	Malay	M	33	30	241	326	1 0	PZI 28	9	—	good	fair	nil	nil	200 000	random blood sugar 156 mg %
61	935/56	Ind	M	61	55	—	—	1 0	SI 52 PZI 28	9	5 yrs	good	fair	anorexia	nil	—	
62	N989	Ind	M	45	42½	184	302	0 5	IZS(SL)40 IZS(L)84	11	18	good	good	initial giddiness	nil	140 000	random blood sugar 139 mg %
63	1013/57	Malay	M	34	33	180	280	0 5	IZS(L)56	11	11½	good	fair	constipation	nil	195 000	random blood sugar 77 mg %
64	N497	Malay	F	51	48	303	454	0 25	IZS(L)32	9	36	good	good	nil	nil	165 000	—
65	1007/56	Ind	M	67	60	120	255	0 75	PZI 52	8	18	good	good	constipation	nil	150 000	—
66	939/56	Ind	F	42	33	147	326	1 0	IZS(L)72	5	39	fair	poor	nil	nil	—	—
67	1819/56	Ind	F	59	57	200	332	1 0	PZI 24	5	5	fair	good	nil	nil	140 000	—

* Key Ind Indian Ch Chinese
† In glucose tolerance test

patients thrice daily—before breakfast and after lunch and dinner. Patients were given full clinical examination whenever they complained of any symptoms that might be construed as probable side effects of the drug, and they were questioned directly for any symptoms referable to the cardiovascular, digestive, urinary, and neurological systems and about any skin reaction.

Patients who were on insulin or tolbutamide, regardless of the quality of control achieved, had their insulin or tolbutamide discontinued for a variable

TABLE 6
CORRELATION BETWEEN AGE AT ONSET OF DIABETES AND CONTROL ACHIEVED

Quality of control	No. of patients per age group*									Total
	26–30	31–35	36–40	41–45	46–50	51–55	56–60	61–65	66–70	
Good	1 (50)	6 (85.7)	6 (60.0)	9 (75)	13 (85.6)	9 (69.2)	3 (75)	1 (50)	1 (50)	49 (73.1)
Fair	1 (50)	1 (14.3)	3 (30)	2 (16.6)	1 (7.2)	2 (15.4)	1 (25)	nil	nil	11 (16.4)
Poor	nil	nil	1 (10)	1 (8.4)	1 (7.2)	2 (15.4)	nil	1 (50)	1 (50)	7 (10.5)
Total	2 (100)	7 (100)	10 (100)	12 (100)	15 (100)	13 (100)	4 (100)	2 (100)	2 (100)	67

* *Figures in parentheses are percentages of total.*

period until glycosuria indicated a relapse of the diabetic state. Chlorpropamide was then given in place of insulin or tolbutamide.

Dosage of chlorpropamide. As the full toxicology of chlorpropamide in man is not known, it was decided to start treatment with the drug in a relatively small dosage given once or twice daily for the first week. Initially, 0.5 gm. of the drug was given, but this was reduced to 0.25 gm., the smaller dose being less likely to produce side effects. The dosage of 0.25 gm. was increased at weekly intervals to 2, 3, or even 4 times daily until control was achieved or until side effects necessitated the discontinuance of treatment. Once control was established, the dosage was reduced to the maintenance level, as adjudged by the relative absence of glycosuria.

Classification of clinical data. Taking into consideration the renal threshold for glucose in the individual patient, the quality of control of the diabetes was assessed on the basis of the degree of glycosuria present. A condition with no sugar in the urine, or only infrequent traces of sugar was designated good. Where traces of sugar in the urine occurred frequently, after dietary deviations had been excluded, the control of the diabetes was classified as fair. Greater glycosuria constituted poor control. The clinical data are presented in TABLES 3, 4, and 5.

TABLE 7
CORRELATION BETWEEN SEX AND RACE AND CONTROL ACHIEVED

Quality of control	Sex		Race				Total
	Male	Female	Chinese	Indian	Malay	Others	
Good	34 (80.9)	15 (60)	16 (64)	23 (74.2)	7 (87.5)	3 (100)	49 (73.1)
Fair	4 (9.5)	7 (28)	4 (16)	6 (19.4)	1 (12.5)	nil	11 (16.4)
Poor	4 (9.6)	3 (12)	5 (20)	2 (6.4)	nil	nil	7 (10.5)
Total	42 (100)	25 (100)	25 (100)	31 (100)	8 (100)	3 (100)	

* Figures in parentheses are percentages of total

TABLE 8
CORRELATION BETWEEN DOSAGE OF CHLORPROPAMIDE AND CONTROL ACHIEVED

Quality of control	No of patients per chlorpropamide dosage gm *					Total
	0.25	0.5	0.75	1.0	1.5	
Good	7 (10.4)	17 (25.4)	14 (20.9)	11 (16.4)	nil	49 (73.1)
Fair	nil	3 (4.4)	4 (6)	4 (6)	nil	11 (16.4)
Poor	nil	1 (1.5)	2 (3.0)	3 (4.5)	1 (1.5)	7 (10.5)
Total	7 (10.4)	21 (31.3)	20 (29.9)	18 (26.9)	1 (1.5)	67 (100)

* Figures in parentheses are percentages of total

Analysis of clinical data Analysis of the clinical data reveals that of the 67 patients treated with chlorpropamide, 49 (73.1 per cent) showed the diabetes (glycosuria) under good control In 11 patients (16.4 per cent) the control was fair, and in 7 (10.5 per cent) the control was poor

Correlation between the quality of control achieved under treatment and any one of the following factors—age at onset of diabetes, race, sex and dosage of chlorpropamide used—is given in TABLES 6, 7, and 8

Discussion

Hypoglycemic activity of chlorpropamide. There is no doubt that chlorpropamide is an effective hypoglycemic agent, as it was able to reduce glycosuria to a minimum in 73.1 per cent of the cases studied. This segment is made up of the 40 good control patients, whose ages at the onset of diabetic symptoms ranged from 30 to 66 years. The degree of control of the diabetes, when related to the ages of the patients at the onset of diabetic symptoms (TABLE 6) does not reveal with any certainty that the drug is more effective in the older than the younger age group, 85.7 per cent of patients between the ages of 31 and 35 achieved good control of glycosuria, while 75 per cent between the ages of 56 and 60 also had good control.

TABLE 9
CORRELATION BETWEEN FASTING BLOOD SUGAR AND CONTROL ACHIEVED

Quality of control	No. of patients per fasting blood sugar levels, mg. per cent*		
	50-150	151-250	251-350
Good	5 (55.6)	29 (76.3)	7 (77.7)
Fair	2 (22.2)	5 (13.1)	2 (22.3)
Poor	2 (22.2)	4 (10.6)	nil
Total	9 (100)	38 (100)	9 (100)

* Figures in parentheses are percentages of total.

The data of TABLE 7, where the sex of the patients is related to the degree of control of the diabetes, shows that 80.9 per cent of male and 60 per cent of female patients achieved good control. It would seem that chlorpropamide is somewhat more effective in male than in female diabetics, but a definite pronouncement on this problem cannot be made without further investigation in a larger series of cases.

In relating the ethnic groups of patients to the number of cases with good control of glycosuria, it is obvious that chlorpropamide is equally effective in Chinese, Indians and Malays.

Chlorpropamide dosage. The dosage of chlorpropamide required to maintain good control ranged from 0.25 gm. to 1.0 gm. daily, with an average total daily requirement of 0.647 gm.

The majority (31 patients) of good-control cases required a maintenance dose of 0.5 to 0.75 gm. daily, these cases constituting 46 per cent of the entire series and 63 per cent of all good-control cases. With the exception

of 1 patient in the poor control group, to whom a daily total dose of 1.5 gm of chlorpropamide was given it is noteworthy that the total daily drug dosage in the fair-control and poor control groups ranged from 0.5 to 1.0 gm

It may be that further increase in the drug dosage for these two groups will reduce further the glycosuria of a few patients

Correlation studies between the fasting blood sugar level and the quality of control achieved under treatment do not indicate that patients with a high

TABLE 10
CORRELATION OF FASTING BLOOD SUGAR TO CHLORPROPAMIDE DOSAGE IN GOOD-CONTROL CASES

Chlorpropamide dosage gm	No of patients per fasting blood sugar levels mg per cent*			Total
	50–150	151–250	251–350	
0.25	1 (14.2)	3 (11.1)	2 (28.4)	6
0.5	4 (57.4)	9 (33.3)	3 (43.2)	16
0.75	1 (14.2)	9 (33.3)	1 (14.2)	11
1.0	1 (14.2)	6 (22.3)	1 (14.2)	8
Total	7 (100)	7 (100)	7 (100)	41

* Figures in parentheses are percentages of total

fasting blood sugar level necessarily required higher dosage to attain good control of their glycosuria Neither does a low fasting blood sugar level indicate a low-dosage requirement to obtain good control Thus the fasting blood sugar range of good control cases is 86 to 350 mg per cent, and 27 of these 41 patients (65.8 per cent) required a maintenance dosage of 0.5 to 0.75 gm daily Correlation between the fasting blood sugar level and the quality of control achieved under treatment is shown in TABLE 9 the relationship between the fasting blood sugar level and the dosage of chlorpropamide required to achieve good control is shown in TABLE 10

Patients previously on tolbutamide therapy In Group *B* (TABLE 4) of this trial series 25 patients were treated with chlorpropamide after glycosuria returned or became more severe when tolbutamide therapy was discontinued In this group 9 patients (36 per cent of the group) were subsequently better controlled on chlorpropamide than on tolbutamide and 4 (16 per cent) were not as well controlled as with tolbutamide The remaining 12 patients (48 per cent) had as good control with chlorpropamide as with tolbutamide

Included in the 4 cases (Nos 28, 32, 41, and 44) in which chlorpropamide was less effective than tolbutamide was 1 patient (No 28) who was given chlorpropamide for 2 weeks, at which time he refused to continue treatment with the drug because of side effects Control was subsequently re established with tolbutamide

A comparison of the dosage of chlorpropamide with that of tolbutamide in patients whose glycosuria was controlled to a similar degree shows the average dose to be 0 66 gm for chlorpropamide and 2 0 gm for tolbutamide Thus weight for weight chlorpropamide is a more effective hypoglycemic agent than tolbutamide

Patients previously on insulin therapy Patients on insulin therapy before chlorpropamide treatment constitute Group C (TABLE 5) of this series Of the 15 patients of this group, 9 (60 per cent) achieved substantially better control of their diabetes than on insulin treatment The insulin dosage ranged from 28 to 136 U daily, with an average of 63 5 U, the average dose of chlorpropamide was 0 83 gm In 4 patients (26 6 per cent) chlorpropamide did not reduce the glycosuria any more than did the insulin given previously In the remaining 2 patients (Nos 53 and 67) treatment with chlorpropamide resulted in poorer control than with insulin In 1, the chlorpropamide dosage was increased to 1 5 gm daily while, in the other, the dose was increased to 1 0 gm daily In both cases, the chlorpropamide was subsequently withdrawn and insulin resumed, good control was re established

Side effects of chlorpropamide Throughout the study with chlorpropamide, a close watch was kept for any side effects that might be considered as due to the drug action Of the 67 trial cases, 39 (58 2 per cent) had no side effects of any description, even on direct inquiry Five patients (7 5 per cent) reacted strongly to the drug (Nos 17, 28, 41, 44, and 53), and severe giddiness, fatigue, anorexia, constipation, nausea, and vomiting necessitated discontinuance of chlorpropamide The remaining 23 patients (34 3 per cent) had mild side effects most of which disappeared spontaneously even when the drug was continued, frequently with increased total daily dosage The distribution of the mild symptoms complained of by the 23 patients (some of whom complained of more than one symptom) follows

Symptoms	Number affected
Constipation	10
Giddiness	7
Skin rash	4
Anorexia	4
Fatigue	2
Epigastric pain	1
Profuse sweating	1

Constipation was the most frequent complaint and apart from a decrease in the frequency of bowel action, patients stated that evacuation seemed incomplete and unsatisfying A few resorted to laxatives but in the majority the constipation declined gradually

Giddiness or dizziness was a not infrequent complaint soon after therapy was started The symptom was not accompanied by others referable to

hypoglycemia it was not dispelled by food and there was no hypoglycemia found upon blood sugar analysis There was some suggestion that the size of the initial drug dosage contributed to the occurrence of the dizziness

Of the 4 patients in whom skin rashes developed (Nos 32 34 35 and 46) 3 had scattered symmetrical mildly itching erythematous eruption which took about 5 to 7 days to subside This erythematous rash did not reappear in the same patient even when the drug dosage was increased subsequently In the fourth patient the rash was purpuric and fairly profuse and a thrombocyte count done 3 to 4 days after the purpuric spots and patches appeared registered 180 000 per cubic millimeter of blood This rash also subsided gradually without reappearance

Hematology and urinalysis There was no appreciable change in the RBC and WBC counts in the patients during treatment with chlorpropamide Thrombocyte counts were done on 45 patients at intervals in 8 patients (22 2 per cent) the counts registered values at and below 100 000 cu mm of blood but none suffered from bleeding The majority comprising 31 cases (68 9 per cent) had counts ranging from 100 000 to 200 000/cu mm of blood The remaining 6 cases (17 8 per cent) had counts higher than 200 000 cu mm It seems that chlorpropamide can produce a fairly persistent reduction in the thrombocyte count although in the single patient who had a purpuric rash (No 34) the count was 180 000 cu mm of blood

No abnormality was found when the urine of patients on chlorpropamide therapy was repeatedly examined at intervals for albumin RBC WBC casts, and crystals

Conclusions

Chlorpropamide is an effective hypoglycemic agent and in this respect it produced good control of the diabetes in 73 1 per cent of the cases studied

Chlorpropamide is as active in the younger patients (31 to 35 years) as in the older (56 to 60 years)

There is some evidence that the drug is more effective in male diabetics than in female diabetics

The hypoglycemic action of chlorpropamide is the same regardless of the racial characteristics of the patients studied

Weight for weight chlorpropamide is a more efficient hypoglycemic drug than tolbutamide 0 66 gm of chlorpropamide being required for maintenance of good control as against 2 gm of tolbutamide

There is no indication that to effect good control of the diabetes chlorpropamide dosage need be proportional to the height of the fasting blood sugar level

The maintenance dosage of chlorpropamide is between 0 5 and 0 75 gm per day

In this study of patients with sthenic plethoric bodily configuration, chlorpropamide treatment yielded a higher percentage and better quality of control of the diabetes than did insulin therapy

Chlorpropamide may produce undesirable side effects which are frequently mild and subside spontaneously

There is evidence that chlorpropamide has a mild to moderate thrombocytopenic action

Acknowledgment

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CLINICOTHERAPEUTIC EV[illegible] [illegible]ROPAMIDE A NEW SU[illegible] THE TREA[illegible]S

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Early pharmacological work indicated that chlorpropamide had a higher potency than other sulfonylurea derivatives namely carbutamide and tolbutamide The objective of the study reported here was to confirm such findings from a clinical point of view in human patients with diabetes mellitus

The diabetic patients in whom chlorpropamide was assayed were laborers of the National Railways of Mexico almost all were traveling workers and for that reason required ambulatory treatment Only patients with uncomplicated diabetes were chosen and none was alcoholic Selection was made regardless of the activity of the diabetic process onset of the disease age sex or constitution of the patient This was done in order to compare the results with those reported in using the earlier antidiabetic drugs which had proved suitable for treating overweight middle aged patients who had mild or moderately active diabetes and were free to develop acidosis [1, 6, 10, 14]

Periodic control during the treatment was performed in 33 subjects for 2 to 6 months control was observed by daily glycosuria tests (3 times a day) and routine clinical and laboratory examinations Of the 33 cases studied 19 were submitted to special weekly laboratory tests including urine analysis blood chemistry RBC WBC and liver function tests These tests were chosen in order to detect changes reported as occurring during treatment with carbutamide or tolbutamide (such changes as a decrease in hemoglobin concentration agranulocytosis and alterations of the RBC or WBC as described in experimental and clinical works) [6, 9, 12] Liver damage is probably the most important complication of the treatment with oral antidiabetic drugs [7, 8] for this reason in several patients a battery of liver function tests was done including thymol turbidity thymol flocculation cephalin flocculation and serum bilirubin The routine blood chemistry tests included those for glucose urea and cholesterol as well as a glucose tolerance test which was done in some patients

The amount of chlorpropamide to be given to each patient was calculated as follows When no previous treatment had been given 1 gm of the drug was allowed in patients with glycemias from 250 to 350 mg per cent when insulin had been administered 40 to 60 U were considered equivalent to 1 gm of chlorpropamide when oral sulfonylureas constituted the previous treatment 1 gm of chlorpropamide was administered instead of 1 5 gm of the other drug Chlorpropamide was used at a dosage of 1 5 gm only in patients who had been treated with more than 60 U of insulin daily or when a combination of insulin plus oral hypoglycemic drugs was necessary

The evaluation of the results was made according to the grade of the diabetic process as well as by comparison of the laboratory analyses performed before and after treatment with chlorpropamide

Clinical Results

The clinical results were classified excellent when complete insulin substitution was achieved with satisfactory control and no observed complications good when satisfactory control was achieved without complete insulin substitution or when temporary intolerance was observed and unsatisfactory when a persistent intolerance was present or when insulin requirements could not be decreased

Of the 33 patients (2 females and 31 males 33 to 69 years old) only 5 had severe diabetes and 3 were underweight (TABLE 1) Twenty were designated as giving excellent results 10 good results and only 3 unsatisfactory results

The hypoglycemic effect of the drugs was observed in every patient 2 had hypoglycemia (demonstrated by blood glucose determination) and many showed temporary intolerance especially at the beginning of the treatment The latter symptoms which disappeared with decrease of the drug dosage were probably due to an early and strong hypoglycemic effect In the former 2 cases of hypoglycemic precoma glucose was administered intravenously but the release of the symptoms was observed only after the drug had been discontinued several hours

The final maintaining doses were 125 mg in 4 patients 250 mg in 6 patients 500 mg in 9 patients 750 mg in 1 patient and 1 gm in 10 patients Only in 3 unsatisfactory cases were the doses increased to 2 5 gm a day before discontinuation In 5 patients the drug was discontinued and neither glycosuria nor hyperglycemia has been observed during 1 to 2 months

Insulin was substituted completely in 3 patients in whom it had been the previous treatment in 1 of these patients 250 mg was sufficient to fill the former requirement of 80 U of insulin daily In 9 cases of combined treatment (insulin plus tolbutamide) the complete replacement of insulin was achieved in 7 patients (superseding former insulin requirements of 15 to 40 U daily) with chlorpropamide in the same amount as the former antidiabetic drugs There were exceptions in 2 cases in which this was reduced still further from 1 gm of tolbutamide to 0 5 gm of chlorpropamide In the other cases of combined treatment the insulin requirement was reduced to 20 U daily maintaining gram for gram the administration of

ıe previous medıca
ropamide as tolbut

amide in the other 15 cases the average reduction was from 1 11 gm of other sulfonylureas to 0 445 gm of chlorpropamide which represents an approximate 60 per cent reduction In every case the change from tolbutamide to chlorpropamide was made easily

Of the 20 cases of excellent results almost all were overweight patients and none had severe diabetes Only 2 of the 10 cases with good results had

TABLE 1
RESULTS OF TREATMENT WITH CHLORPROPAMIDE

General data						Previous treatment		Treatment with chlorpropamide		
No	Age	Sex	Evolution time (years)	Type of diabetes	Constitution *	Insulin	Oral antidiabetic drugs	Insulin (U day)	Chlorpropamide (gm /day)	Duration of treatment (months)
Excellent										
1	44	M	2	Slight	+10		‡1 00		0 500	1
2	49	M	17	Moderate	Normal		‡1 25		0 250	2
3	50	M	11	Moderate	+10	‡20	1 00		1 000	2
4	54	M	4	Moderate	−20		1 00		0 125	3†
5	54	F	4	Moderate	+20	30	1 00		1 000	2
6	55	M	1	Slight	+20				0 125	1†
7	55	M	5	Slight	+20		1 00		0 500	3
8	56	M	22	Moderate	+20	30			0 500	1
9	57	M	1	Moderate	+10		‡1 00		0 125	6†
10	57	M	4	Moderate	+10		‡1 50		1 000	1
11	60	M	1	Moderate	+10		0 50		0 250	2
12	61	M	6	Moderate	+20	‡30	1 00		1 000	1
13	62	M	5	Moderate	Normal	20	1 00		0 500	1
14	62	M	6	Moderate	Normal		‡1 00		0 125	3†
15	63	M	4	Moderate	+10	40	1 00		1 000	2
16	63	M	5	Slight	+30		1 50		1 000	1
17	63	M	6	Moderate	+20	‡30	1 00		1 000	1
18	63	M	6	Moderate	+20	30			0 500	2
19	66	F	10	Slight	+20		‡1 00		0 250	1
20	69	M	19	Moderate	Normal		1 00		0 500	2
Satisfactory										
21	38	M	3	Moderate	Normal		‡1 50		0 250	2
22	52	M	4	Moderate	+20	‡80			0 250	3
23	54	M	4	Moderate	+20		‡2 00		0 500	1
24	58	M	18	Slight	Normal				0 750	1
25	59	M	14	Severe	Normal	40	1 00	20	1 000	6
26	60	M	4	Moderate	Normal		‡1 00		1 000	1
27	62	M	5	Severe	+20	‡60	1 00	40	1 000	2
28	62	M	20	Moderate	+10		‡1 00		0 500	1
29	68	M	13	Moderate	+20	‡15	1 00		0 500	6†
30	68	M	22	Moderate	Normal		‡0 50		0 250	2
Unsatisfactory										
31	33	M	8	Severe	−20	‡60		30	2 500	1
32	52	M	12	Severe	−20	40		40	1 500	1
33	63	M	2	Severe	+10	60		60	1 500	1

severe diabetes and all had a normal weight or were overweight The 3 cases of unsatisfactory results h[illegible]
trolled previously with 80 [illegible]
requirements with as much [illegible]
2, one was underweight, b[illegible]
old), and both were psychopathic These last 2 cases showed persistent

gastrointestinal intolerance to the drug, and no reduction of the insulin requirements could be achieved

The side effects observed during the treatment with chlorpropamide were almost always moderate, with only transitory intolerances, probably due to overdoses of the drug Gastrointestinal disturbances such as hiccough gastric pain, or diarrhea were present in six cases and were of important intensity in only two In every patient the symptoms disappeared with the reduction or withdrawal of the drug Nervous system disturbances such as nausea or vertigo, appeared in two cases, disappearing with the temporary discontinuation of the drug There was only one case of dermatological reaction this consisted of papula on the head of the patient and was a moderate reaction lasting a short time and disappearing with the reduction of the drug dosage Three other patients (one not included in these thirty three cases) observed a generalized peripheral vasodilatation, especially over the face and neck This reaction, of variable intensity, was originated by the ingestion of a cup of wine or glass of beer, the test was made with other patients, but the effect could not be reproduced

Laboratory Data

Unimportant changes were detected by routine urine analysis, no hematuria, cells or casts appeared during administration of the drug A slight albuminuria was present in some of the patients this did not increase significantly in any case and in some, was even lower than in the initial control period of the study This effect was probably due to a better control of the altered metabolic situation

A decrease in red blood cells observed in 7 patients was less than 1 million in each of 6 and, in one case, the decrease was from 5,100,000 to 4,100,000 Microcytic hypochromic anemia was present in 2 patients before treatment and did not show any modification during the time the drug was given, 5 of the 7 patients mentioned showed a tendency to hypochromic anemia In the rest of the cases, the RBC was practically unchanged No important change was found in the WBC or in the total leukocyte or differential counts

Total cholesterol was normal or slightly increased before treatment No important modification could be found in this determination Blood urea, slightly increased (37 to 44 mg per cent) in 6 cases, was normal, after treatment in every case except 3 of the previous 6 mentioned, in these a slight further increase was found (44 to 55 mg per cent)

One to 3 liver function tests were performed before and after treatment on each of the 19 subjects thymol turbidity was done in 16 cases, cephalin flocculation in 15 serum bilirubin in 11 and a Bromsulphalein test in 3 No damage was demonstrated by these liver function tests In 7 cases in which thymol turbidity was slightly increased before the treatment with chlorpropamide (from 6 to 14 U), it was found to be within the normal values after treatment

In the two cases in which glucose tolerance tests were done, the general morphology of the test was practically unchanged before and after treatment

Conclusions

The new compound chlorpropamide showed its hypoglycemic effect in every case Excellent or satisfactory results were observed in 28 cases of moderate or slight diabetes unsatisfactory results were reported for 3 patients with severe diabetes and 2 cases of severe diabetes gave satisfactory results The indications and contraindications of this drug seem to be the same that have been reported for carbutamide and tolbutamide [1]

The toxic effects seem to be similar to those reported for the other anti diabetic drugs the observed lack of response to glucose administration was similar to that reported in animals by Houssay [15]

Although these drugs seem to have no effect on the metabolism of alcohol [16] the ingestion of it produced important peripheral vasodilatation in three cases this effect was not accompanied by hypertension or headache and lasted for two to four hours

No toxic effect could be shown by the liver function tests such findings are difficult to evaluate because of the liver dysfunction that is frequently present in treated or untreated cases of diabetes mellitus [17 19]

The potency of the drug seems to be 2 to 3 times that of carbutamide or tolbutamide but has individual variation from case to case some patients are well controlled with 0 125 gm of chlorpropamide while others require as much as 1 or 1 5 gm in order to achieve correct control

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CLINICAL STUDIES WITH CHLORPROPAMIDE

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Fifteen office patients with diabetes were treated with chlorpropamide In 6 it was necessary to discontinue the drug 4 developed ketosis, 2 lost considerable weight and had poor appetites, although their blood sugars approached normal values and there was no glycosuria, 9 are still under observation The shortest period of follow up was 2 months, the longest, 6 *The ages of the 4 men and 5 women treated ranged from 35 to 70 All* but 1 are diabetics who had been on insulin and who, at other times, managed their diabetes by dietary means, thus providing the factors necessary for a comparison

All of these patients were permitted a normal or usual diet They neither weighed nor measured their food, and they had sweets on occasion At first they were seen every other day, then at weekly intervals and now at monthly visits The dosage of chlorpropamide varied from 500 to 1000 mg At each visit the patients were *questioned as to symptoms of diabetes or* hypoglycemia (or both), pruritus, skin eruptions, or weakness, later they were also questioned as *to loss of appetite Body weights were recorded,*

amined

ade for

counts

At first the immediate response to the drug was tested The 5 patients used were 30 to 60 years old, and had had diabetes from 1 month to 14 years *All came to the office without breakfast and, after blood was drawn* and a urine specimen obtained, 1 gm of chlorpropamide was administered to each One hour later, blood and urine samples were obtained, and the patients were then sent out for a breakfast consisting of fruit juice, 2 eggs, 1 slice of toast, and a beverage, some had cereal instead of the eggs The blood and urines were examined two hours after this breakfast, and a fasting specimen was taken 24 hours after the initial administration of the drug

From TABLE 1 *it is clear that only 2 of the 5 unselected patients showed a* hypoglycemic reaction to the test dose Patient W Mc showed a consistent fall in the blood glucose the effect persisting overnight The chlorpropamide blood levels were constant, and only a slight drop was seen after 24 hours With so definite a response, this patient appeared suitable for therapy with chlorpropamide However, since he was obese and his diabetes was of only 1 month's duration, he was treated by subcaloric diet only, no drug being given *He lost weight slowly and, with this weight reduction,* his glycosuria cleared He has no symptoms and his last fasting blood sugar was 130 mg per cent Had we given him chlorpropamide with the prescribed diet, we certainly would have considered the result a successful

* Chlorpropamide levels were determined by Chas Pfizer & Co Inc, Brooklyn N Y

response to the drug. Patient P. B. has had diabetes for 4 years. His symptoms were inconstant and his glycosuria, although occasional, was most apparent postprandially. He weighed 215 lb. and 4 years ago dietary therapy alone was advised. On a subcaloric diet he lost weight with the loss of 30 lb.; however, he also had symptoms of diabetes with intense fatigue as the most troublesome feature. He refused insulin and chlorpropamide seemed ideal for him. His hypoglycemic response showed the test do e to be a failure even though the drug's blood level was sustained. His firm refusal to use insulin, however, prompted us to try chlorpropamide. He

TABLE 1
DATA ON SIX PATIENTS TREATED WITH CHLORPROPAMIDE

Patient	Sex	Age	Duration of diabetes	NPH Insulin U	Blood sugar, mg per cent*				Chlorpropamide blood levels, µg/ml			
					Fasting	1 hour after 1.0 gm dose	2 hours after breakfast	After 24 hours	Fasting	After 1 hour	After 3 hours	After 24 hours
R. M.	M	50+	1 month	0	215 S ++	192 S +	158 S +	134 S 0	0	94	97	87
R. B.	M	50+	4 years	0	308 S 4+	204 S 3+	286 S 4+	265 S 4+	0	8	93	81
H. J.	M	40+	3 years	10	210 S +	180 S +	104 S 0	153 S +				
J. D.	F	30+	10 years	50	130 S 4+ AC +	330 S 4+ AC 3+	440 S 4+ AC 4+	Terminated R25	0	46	64	—
S. R.	M	60	14 years	60	337 S 4+ AC 0	344 S 4+ AC 4+	395 S 4+ AC 4+		0	33	24	59† 50†
E. C.	F	60	2 years	0	360 S 4+		360 S 4+					

* Lower figure or figures: S represents urine sugar; AC, acetone.
† Results are for same samp e sp

was given 1.0 gm. daily; surprisingly, the glycosuria cleared, most of his symptoms of diabetes disappeared and for 2 months his fasting blood sugars ranged from 170 to 200 mg. per cent. The liver function tests were normal as was the hemogram. The chlorpropamide level was 318 µg./ml. All of this was spectacular but although he ate well, he did not enjoy his food and was losing weight at the rate of 5 lb. per month. Because we suspected some unpleasant complication he was hospitalized but an extensive study failed to reveal any abnormalities. While he was in the hospital the chlorpropamide was continued and he chose his own foods. Although all fractional urine specimens were sugar free and the blood sugars were 125 and 132 mg. per cent, he felt tired, did not enjoy his food and failed to gain weight even though he forced himself to eat his meals. Failure to improve

THE USE OF CHLORPROPAMIDE IN BRITTLE AND POORLY CONTROLLED DIABETES MELLITUS*

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The development of the oral hypoglycemic agents has led to some sweeping revisions in concepts about the control of the abnormal carbohydrate metabolism in diabetes mellitus. With the compounds presently available (tolbutamide in the United States and
oral therapy is available only t
diabetes.[1-3] It is well known th
diet alone or by diet with small doses of insulin. Consequently, use of the oral agents for these patients has been more a convenience than a necessity. Patients in urgent need of some agent that will provide more stable control are the juvenile diabetics and the adult diabetic patients who are poorly controlled on any dose of insulin. Included in the latter group are those patients who are brittle in the sense that there is irregular daily alternation between gross hyperglycemia and overt or occult hypoglycemia. In these patients many factors are involved, such as poor diet, irregular habits, subcutaneous reactions at the sites of insulin injection, insulin overdosage, inability to break down complex insulin, and infection—to name only a few. As a rule, tolbutamide and carbutamide are ineffective in such patients, although an occasional excellent response has led to the hope that some other sulfonylurea would be helpful in achieving significantly improved clinical control.

With the development of chlorpropamide, which was thought to be a more potent agent, it was felt that an adequate trial in the brittle, unstable, and *poorly controlled diabetic was justified. Patients were selected from* the Diabetic Clinic at Jefferson Davis Hospital and referred to a special clinic for oral therapy. The patients attending the diabetic clinic at this hospital have been characterized in detail in the literature.[4] In general they are in poor financial circumstances, of low intelligence, and poorly educated. Most are obese, and urinary tract infection is a major problem.

Chlorpropamide was started in large doses (as much as 1.5 gm. per day) *in the poorly controlled* diabetic receiving large doses of insulin. In general the patient's insulin dosage was reduced by one half when chlorpropamide therapy was instituted. This was the usual practice if the patient was receiving more than 50 U. of insulin. Those with mild, stable diabetes received 500 to 1000 mg. per day as the initial dose and insulin was discontinued. Treatment was carried out on an outpatient basis and the

* The work reported in this paper was supported by a grant from Chas. Pfizer & Co., Inc., Brooklyn, N. Y.

† Trainees in Diabetes Training Grant Program, National Institute of Arthritis and Metabolic Diseases, Public Health Service, Bethesda, Md.

patients were provided with explicit instructions to communicate with one of the physicians on the metabolic service if anything unusual occurred No additional instruction in dietetics was given to any of these patients this was done deliberately to obviate the possibility that any degree of improved control would result from improved dietary habits The patients were checked at weekly intervals until they had become stabilized on the oral therapy, the period between visits was then lengthened Doses of insulin and chlorpropamide were altered as deemed advisable from the clinical course White blood counts, differentials blood urea nitrogens thymol turbidity blood sugar and chlorpropamide blood levels were determined at each visit The patients who missed appointments were asked to appear later Sixty-one patients have received therapy for a time sufficient to allow evaluation

Results

The status of the sixty one patients is summarized in TABLE 1 Three patients died while receiving chlorpropamide therapy One of these died

TABLE 1
STATUS OF PATIENTS OBSERVED IN STUDY

Deaths since chlorpropamide therapy instituted		3
Not related to drug	2	
Related to drug	1	
Received therapy for 1 week for special metabolic study		3
Drug discontinued because of increasing B U N & symptoms		4
Restarted on drug therapy	2	
Lost to follow up		4
Uncooperative drug therapy discontinued		6
Toxic reactions drug discontinued		12
Withdrawn because of poor control		1
Receiving uninterrupted drug therapy		28
Total		61
Presently on therapy		30

of what appeared to be a myocardial infarction following a typical allergic reaction to penicillin Chlorpropamide therapy had been discontinued one week before death and could be connected in no way with the death The second was a patient with advanced generalized arteriosclerosis who developed an extremely severe urinary tract infection and died of septicemia It was felt that chlorpropamide therapy was not involved in this patient s demise The death of the third patient was felt to be related to the chlorpropamide therapy

Case Reports

S K was a 61 year old white female who was diagnosed as having mild diabetes mellitus in 1954 She had a marked aversion to injections and refused to take insulin Control was good but with minor stress she became

poorly controlled She requested oral therapy and was started on 500 mg of chlorpropamide daily in a divided dose Approximately 2 weeks later she became ill and was treated by a private physician who made a diagnosis of respiratory tract infection The patient was given Azotrex which contains tetracycline sulfamethizole and Pyridium The physician found no evidence of hepatitis at the time of his examination Her symptoms became more acute and 4 days later she was admitted to Jefferson Davis Hospital suffering with nausea vomiting dehydration and jaundice She rapidly developed electrolyte disturbances and congestive heart failure and expired At autopsy there was hepatitis of the cholestatic type most commonly seen with chlorpropamide idiosyncrasy This case will be reported in detail elsewhere It is felt that had this patient reported to the clinic at the time she became ill as she had been instructed she probably would have survived since she continued on sulfonylurea therapy from the onset of symptoms until admission to the hospital

Significant improvement in controls was achieved in four patients but the drug was discontinued because of an increasing level of blood urea nitrogen accompanied by symptoms of malaise weakness and lethargy It was originally felt that this might be a consequence of drug therapy but it is now felt that an acute flare up of chronic urinary tract infection caused most of the symptoms Two of the four patients have received chlorpropamide therapy again with no untoward effect Six patients were lost to follow up

Patients who had been actively uncooperative in the therapy preceding that with chlorpropamide often continued this erratic behavior after being started on oral therapy It is our impression that these patients did not do well especially when judged by a sense of well being general appearance and clinical course Often their chemical control was quite good Chlorpropamide blood levels fluctuated quite widely probably indicating that they had taken their tablets erratically It is felt that patients who continue uncooperative and erratic should not be continued on this form of therapy Toxic reactions of such severity as to force abandonment of chlorpropamide therapy occurred in 12 patients 2 showing somnolence 4 dermatitis and 6 gastrointestinal symptoms

It was necessary for the families of 2 patients with somnolence to go to extremes to awaken them to receive food and medicine One patient who developed a skin rash also experienced mild to moderate somnolence on the drug Of the 12 patients who discontinued chlorpropamide because of toxic reaction 6 had received a maximum dose of 1000 to 1500 mg daily and 6 had received a maximum of 250 to 750 mg daily Of the 32 who had discontinued drug therapy at some time 10 had received at least 1500 mg of chlorpropamide for days to weeks while of the 30 presently on therapy, 13 have received 1500 mg or more daily for a week or longer Hypoglycemia was not mentioned in the side effects as this is a natural consequence of the drug therapy with the large doses being given in this study hypoglycemia was a common phenomenon A large percentage of all of the patients treated had mild hypoglycemia some time before they were stabilized

Ten of the patients underwent moderate hypoglycemic episodes while 3 had severe hypoglycemia

Of the thirty patients still receiving chlorpropamide seventeen are in excellent control, three are in good control and ten are in fair control all are significantly improved relative to their condition during previous therapy

Of the original 61 patients 33 may be classified as poorly controlled or brittle diabetics 19 of these are presently receiving chlorpropamide This agent has proved to be extremely useful in improving the control of the *patient who had been uncontrolled on other regimens Chlorpropamide was* used in combination with insulin in 13 of these cases In 5 however, insulin

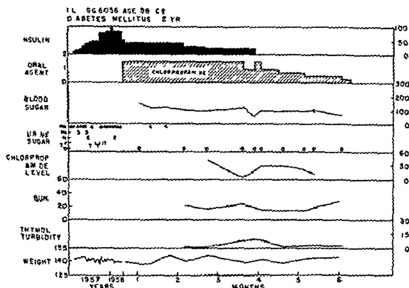


FIGURE 1

was eventually discontinued chlorpropamide was used alone in patients who had received as much as 90 U of insulin The following case reports illustrate the results obtained

I L a 58 year old Negro female in whom the diagnosis of diabetes mellitus was made in 1956 was admitted to the hospital in September 1956 with a subacute myocardial infarction and congestive heart failure She was treated with digitalis, diuretics, bed rest and diet with good results Her course since that time is shown in FIGURE 1 She developed an almost constant 4 plus glycosuria associated with general lethargy and malaise and with *occasional episodes of overt hypoglycemia The response to chlor*propamide has been quite good Insulin has been discontinued and the *patient is maintained on 250 mg of chlorpropamide daily*

B S is a 63 year old Negro female with diabetes mellitus of 22 years *duration* When first seen at the hospital in *1939* she had a urinary tract

infection associated with chronic cervicitis In 1944 she was treated for a Bartholin abscess and, in 1953, she was found to have a peripheral neuropathy and atypical arthritis In 1957 a diagnosis of arteriosclerotic heart disease *with auricular fibrillation and congestive heart failure* was made There was a satisfactory response to digitalis, diuretics, and salt restriction This patient has a seasonal job and finds it difficult to obtain adequate food when unemployed in the summer When this occurs she discontinues her hypoglycemic therapy, as shown in FIGURE 2 She is now receiving 500 mg chlorpropamide daily, which provides more adequate control than as much as 100 U of insulin This patient was hospitalized on August 26, 1958, and

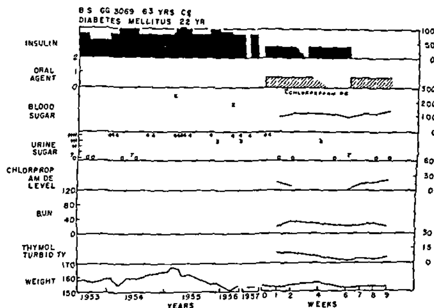


FIGURE 2

a small granuloma of the lung resected She was treated with chlorpropamide throughout her hospital stay, with the exception of the day after surgery, on which she received no hypoglycemic therapy In the immediate postoperative period moderate glycosuria appeared, mainly while she received intravenous therapy *On the third postoperative day she received* 30 U crystalline insulin in divided doses The glycosuria cleared the next day and she was discharged in excellent condition 1 week later This patient is achieving significantly better control on chlorpropamide alone than she did on insulin alone In addition, it was possible to carry her

duration, was first seen at Jefferson Davis Hospital in 1951 with uncontrolled diabetes mellitus, mild acidosis, and pyelonephritis She was admitted

1 month later with a carbuncle on the right buttock acidosis and severe pyelonephritis In the period 1952 to 1956 she was admitted 5 times with severe urinary tract infection usually associated with acidosis It was possible to suppress the urinary tract infection but not to eradicate it in spite of all diagnostic and therapeutic procedures In 1957 she was found to have a peripheral neuropathy and Kimmelstiel Wilson syndrome was

all forms of antibiotic therapy In spite of the infection it was possible

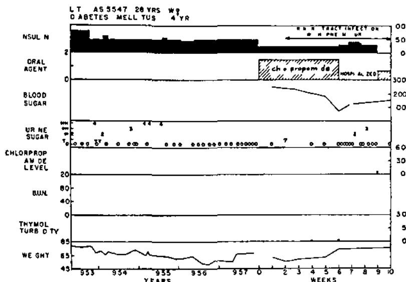


FIGURE 3

to discontinue the use of insulin and to achieve adequate control with 750 mg of chlorpropamide daily Dissatisfied because of the continued urinary tract infection she did not cooperate with the oral hypoglycemic therapy and began to manifest evidence of poor control This patient is classified in our uncooperative group of patients It was evident that as long as she cooperated with therapy her control (both chemical and symptomatic) remained quite good when she became uncooperative she rapidly developed symptoms that required discontinuation of chlorpropamide and reinstitution of insulin This patient s history is summarized in FIGURE 3

S W a 30 year old Negro male in whom the diagnosis of diabetes mellitus had been made four years previously had been hospitalized for two weeks in January 1955 with poor control and acidosis He had previously failed to respond to carbutamide and tolbutamide All general manifestations of poor control were present such symptoms as weight loss weakness inability

to carry out sustained effort, and increasing susceptibility to infections The response to chlorpropamide in this patient has been excellent As in this patient, it is easy to see that chlorpropamide is effective when others of the sulfonylurea compounds have proved ineffective This history is shown in FIGURE 4

F D, a 55-year-old white female with known diabetes of two years' duration was first seen at the hospital in May 1955 with paroxysmal auricular tachycardia that responded to parenteral digitalis She was hospitalized for four weeks in December 1956 because of poor diabetic control, angina pectoris and a posterior myocardial infarction of indeterminate age During

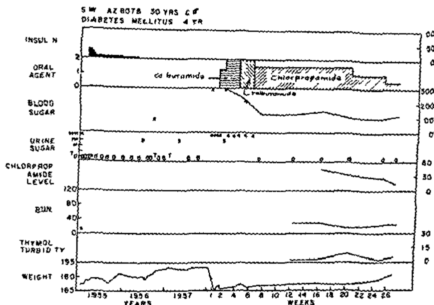


FIGURE 4

this hospitalization tolbutamide therapy was started but was discontinued with the onset of an acute severe urinary tract infection Later she was treated with carbutamide with fair initial response but secondary failure Initial insulin therapy produced almost no improvement When the insulin dosage was increased the patient developed rather severe hypoglycemia at irregular intervals She developed severe hypoglycemia on the first day of combined therapy of chlorpropamide and insulin Reduction in the chlorpropamide dosage led to significantly improved control This patient subsequently left the community, and recent communication with her private physician at her new location gave the information that she has remained in excellent control. Data on this history are in FIGURE 5

A N is a 59-year-old white female whose primary difficulty has been extensive nodule formation at the sites of insulin injection As a consequence, she has alternating periods of hyper and hypoglycemia that, at

times, have completely disrupted her life With chlorpropamide therapy, the dose of insulin given has been reduced significantly the nodule formation has not been a major problem at this reduced dose level of insulin Details of this history are shown in FIGURE 6

B C is a 66 year old white female with diabetes of thirteen years' duration and has been observed for more than twenty years at the Jefferson Davis Hospital This patient has myxedema and has had surgical removal of a nontoxic benign thyroid adenoma For several years she has had recurrent acute episodes of a chronic severe urinary tract infection She has arterio sclerotic heart disease with a left bundle branch block and peripheral neu-

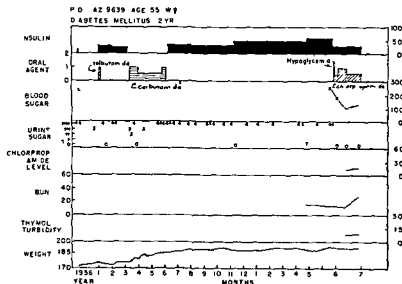


FIGURE 5

ropathy At no time has this patient's diabetes come under satisfactory chemical control, in spite of all efforts in the Outpatient Clinic, plus multiple hospitalizations The response to chlorpropamide has been dramatic, however, she continues with urinary tract infections, and the weight loss noted in the chart is due to the continued infection rather than the chlorpropamide therapy This case is illustrated in FIGURE 7

L W, a 70 year-old white male with diabetes mellitus of 20 years' duration, was admitted to Jefferson Davis Hospital in 1950 with acidosis, and *in 1953 with the same complaint In 1957* he was found to have arterio-sclerotic heart disease with congestive heart failure, which responded to digitalis, diuretics, and salt restriction At the time of this hospitalization he also had mild hypertension and bronchopneumonia He has received as much as 130 U of insulin daily, with a clinical course characterized by alternating hyper and hypoglycemia He refuses to take less than 100 U

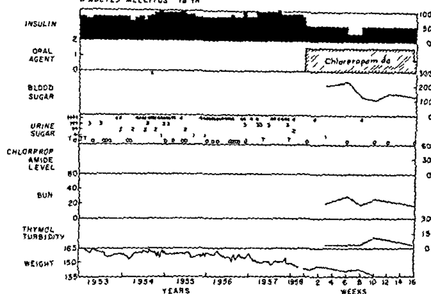


Figure 6

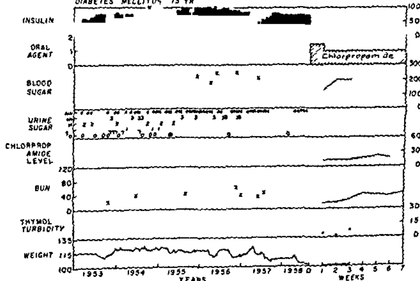


Figure 7

of insulin a day, however, the addition of chlorpropamide to 1500 mg daily has significantly improved his chemical control. His clinical course is much smoother as he does not alternate between hyper- and hypoglycemia and feels much better than he has for many years. Details are shown in FIGURE 8.

J. J. is a 35-year-old Negro female with diabetes mellitus of 7 years duration. At onset her diabetes was relatively mild and comparatively easily controlled. In the past two years, however, she has presented one of the most unusual and bizarre problems in diabetic control that has ever been seen in this clinic. She has developed extensive nodule formation at the sites of insulin injections and, in addition, approximately once a month

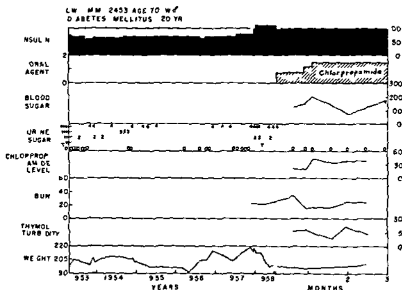


FIGURE 8

develops a delayed hypersensitivity response at the sites of injection. During these two years it has never been possible to achieve significantly improved control in spite of the fact that it has been possible to put her into an environment that is well controlled, where she receives meals regularly and where nutritional deficiencies are corrected. She was first started on DBI. By chemical criteria this produced only minimal improvement in control; by clinical criteria, however, the results were quite striking. It was then felt desirable to change from DBI to chlorpropamide to determine if any improvement in the chemical control could be achieved. As may be seen in FIGURE 9, there was very little difference in the control achieved by each of these drugs. It should be noted, however, that she is receiving about 50 per cent of her previous dose of insulin. It is obvious, too, that occasionally she exhausts her insulin supply and neglects to return to the hospital for an additional supply. As a criterion for the improvement in control in this patient it

may be stated that in the 1½ years prior to the institution of oral therapy, this patient had been hospitalized one fourth of the time Since the institution of oral therapy she has not been hospitalized

W A is a 64 year-old white male with diabetes mellitus of three years' duration When the diagnosis was first established, he was treated with small doses of insulin However, he failed to return to the clinic and was not seen again until early in 1958

Because of the previous good results of chlorpropamide it had been the desire of the metabolic section to attempt to treat a person in acidosis with intravenous chlorpropamide On admission, this patient was in moderate

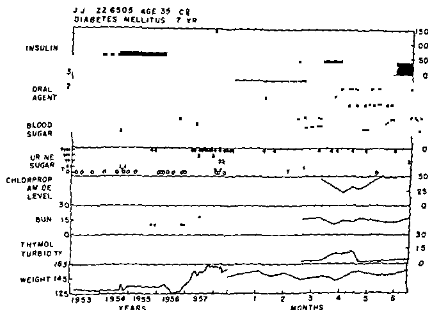


FIGURE 9

acidosis with clinical signs and symptoms, but he was not in coma He was administered 1000 mg of chlorpropamide intravenously during a 6 hour period Other fluids were given as is indicated in FIGURE 10 It is evident that the patient s acidosis was relieved promptly Glycosuria and hyperglycemia continued under the impact of the intravenous glucose A total of 70 U of insulin was given after the patient was cleared of acetonuria, ... attempt to decrease the glycosuria than to control the acidosis

derate acidosis may be treated with

cement, without the administration

patient indicates that the use of

be worth further exploration

As may be seen practically all the patients received large doses of chlorpropamide for weeks to months They tolerated these large doses quite well, and it is our impression that the only toxic manifestations that occurs

more frequently in this group of patients are nausea and vomiting It appears then that the poorly controlled diabetic receiving large doses of insulin will tolerate large doses of chlorpropamide for long periods of time without manifesting overt toxicity

The term diabetic control has many connotations to many people Control may be spoken of as clinical or chemical with subdivisions as far as glycosuria hyperglycemia weight gain sense of well being and other criteria are concerned In the 30 patients who are presently on therapy an attempt was made to subdivide diabetic control into control of glycosuria control of blood sugar control as judged by weight gain or loss and control as

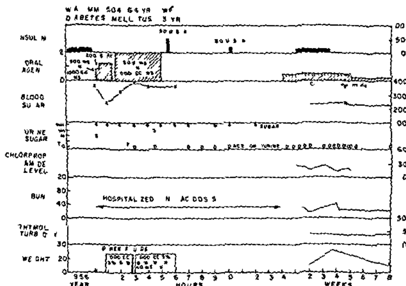


FIGURE 10

judged by general clinical course Degrees of control in each category were expressed as excellent good fair poor and uncontrolled As judged on the basis of glycosuria control was excellent in 20 good in 3 fair in 5 and poor in 2 Based on maintenance of a satisfactory postprandial blood sugar excellent control was achieved in 14 good in 7 fair in 6 and poor in 3 On the basis of weight 25 were in excellent control 3 good 1 fair and 1 poor Symptomatically 26 were in excellent control 3 good and 1 fair It is noteworthy that the drug was discontinued when patients were uncontrolled or in poor control symptomatically

In addition to the brittle and poorly controlled diabetics reported above there were 8 patients in whom the diagnosis of diabetes mellitus had been made recently All of these were of the type that needed some hypoglycemic therapy and chlorpropamide was administered Of these 8 7 are still

receiving oral therapy and only one is receiving this in combination with insulin

Summary

Chlorpropamide has proved to be a valuable adjunct to the therapy of brittle and poorly controlled diabetics In patients receiving large doses of insulin it has been possible in most instances to reduce the dose of insulin by at least 50 per cent and to achieve a significantly smoother and more stable course for the patient One case of toxic hepatitis with death is reported apparently this was due to idiosyncrasy to the drug Chlorpropamide remains effective under such stress situations as infection and surgical operation and on the basis of one case in the presence of acidosis

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Discussion of the Paper

GARFIELD G DUNCAN (*Philadelphia Pa*) I have three patients who became refractory to tolbutamide therapy and later required a larger dose of insulin than previously Have any of the investigators in basic research repeated the experiments of Allen who performed a partial pancreatectomy in dogs to produce a state just short of diabetes and then provoked diabetes by overfeeding thus stimulating the islets of Langerhans?

It seems to me that there may exist a similarity between these experiments and experiences with the patients who require more insulin than before sulfonylurea treatment I suggest that we do the same thing and try to precipitate diabetes in such partially pancreatectomized animals with the sulfonylurea compounds Although this may have been done I have not heard of it

MARTIN G GOLDNER (*Jewish Chronic Disease Hospital Brooklyn N Y*) The question whether secondary failures to sulfonylureas have an increased insulin requirement is an important one From what I have seen and from what I know of the literature Lamar s experience does not appear to be the rule I recall for instance Pfeiffer s[1] series of thirty secondary failures only three or four of these had an increased insulin requirement and the increase was rather small while all others required the same amount or even less than that needed prior to sulfonylurea treatment However one must realize that there are at least three different types of so called secondary sulfonylurea failures there is first the patient who overeats and relaxes his diet when placed on the drug second the true secondary failure who without any detectable cause escapes from control and third the patient in whom the diabetes itself has become worse We must not forget that we deal with a disease that may be stable for some time but that may also progress at any

time It would not be surprising if the first or the third type need more insulin but neither would indicate true failure of the sulfonylureas The first is a failure of dietary management the third an idiopathic aggravation of the disease Only the second group can be linked causally to the hypoglycemic drugs and in this type the general experience seems to be that the insulin requirement is practically unchanged

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ADVANTAGES AND DISADVANTAGES IN SHIFTING PATIENTS FROM TOLBUTAMIDE TO CHLORPROPAMIDE

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Because of the chronicity of diabetes mellitus, permanent continuous treatment is necessary Therefore, efforts have been made (and continue to be made) to develop new clinically effective agents that have pronounced hypoglycemic effect It is most desirable that minimal dosage of such agents produce an optimal antidiabetic effect and, further, that the lowest possible incidence of untoward side reactions be associated with therapy In keeping with this research objective, the Austrian branch of Pfizer International Inc, New York, N Y, supplied us with the new antidiabetic drug, chlorpropamide, for clinical evaluation

Materials and Methods

Two hundred inpatients were treated with chlorpropamide, special consideration being given to evaluating the clinical advantages and disadvantages of the drug in comparison with those of tolbutamide and carbutamide

TABLE 1

DATA ON DIABETICS SUCCESSFULLY CONTROLLED WITH CHLORPROPAMIDE

Previous measures	No of patients	No with good control on chlorpropamide only
Diet only	45	43
Tolbutamide and carbutamide	26	20
Insulin and introductory chlorpropamide	56	40
Prolonged insulin	73	43
Total	200	146

More than 1700 diabetic patients have been treated with the latter 2 preparations for a period of almost 3 years In addition, 15 outpatients were put on chlorpropamide during our clinical investigation

In July 1958 we were able to report on 136 patients studied to that time We have now enlarged this group to 200 patients who either had been on insulin or whose diabetes had not been satisfactorily controlled by dietary restriction alone, even though they were hospitalized The above figure also includes those patients who had been treated with tolbutamide, carbut... than ... thirds of

insulin, the remaining 83 had been ... Of the latter, 40 received insulin ...

admission to the hospital in order to stabilize their diabetic condition rapidly the remaining 43 had been controlled with insulin for more than 10 years

The protracted therapy employed for periods as long as 5 months (follow ups were carried out at 3 week intervals) brought about very good metabolic equilibrium in 25 of 38 cases and just sufficient control in 5 cases In the latter fasting blood sugar was about 200 mg per cent and 24 hour glycosuria were up to 15 gm *In the remaining 8 patients control with chlorpropamide* was discontinued because of increasing glycosuria which obviously

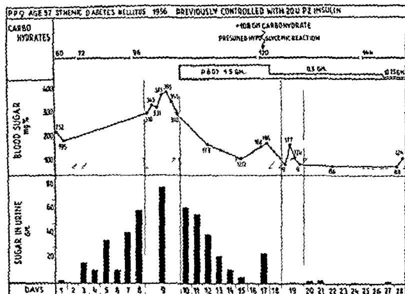


FIGURE 1 Chlorpropamide induced increase in carbohydrate tolerance

resulted from neglect of the strict dietary rules Acidotic and pregnant diabetics were not treated with chlorpropamide The usual complications for example retinopathy sclerosis of the coronary arteries, nephropathy and obliterating arteriosclerosis of the extremities do not constitute contraindications for chlorpropamide treatment It follows that the control of diabetes with chlorpropamide is more successful—the majority of the patients respond in those in whom diabetes becomes manifest at or after the fifth decade of life This applies especially to diabetics who previously were controlled by dietary restriction alone (TABLE 1) In such cases chlorpropamide allows a larger intake of carbohydrates while effecting a nearly normal fasting blood sugar an excellent 24 hour blood sugar profile and sugar free urine (FIGURE 1)

Also evident is the fact that in patients taking insulin, the possibility of success with chlorpropamide decreased as the need for insulin increased In *the majority of cases requiring less than 40 U insulin daily chlorpropamide*

ADVANTAGES AND DISADVANTAGES IN SHIFTING PATIENTS FROM TOLBUTAMIDE TO CHLORPROPAMIDE

J Bloch and A Lenhardt

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Because of the chronicity of diabetes mellitus permanent continuous treatment is necessary Therefore efforts have been made (and continue to be made) to develop new clinically effective agents that have pronounced hypoglycemic effect It is most desirable that minimal dosage of such agents produce an optimal antidiabetic effect and, further that the lowest possible incidence of untoward side reactions be associated with therapy In keeping with this research objective the Austrian branch of Pfizer International Inc New York N Y supplied us with the new antidiabetic drug chlorpropamide for clinical evaluation

Materials and Methods

Two hundred inpatients were treated with chlorpropamide special consideration being given to evaluating the clinical advantages and disadvantages of the drug in comparison with those of tolbutamide and carbutamide

TABLE 1
DATA ON DIABETICS SUCCESSFULLY CONTROLLED WITH CHLORPROPAMIDE

Previous measures	No of patients	No with good control on chlorpropamide only
Diet only	45	43
Tolbutamide and carbutamide	26	20
Insulin and introductory chlorpropamide	56	40
Prolonged insulin	73	43
Total	200	146

More than 1700 diabetic patients have been treated with the latter 2 preparations for a period of almost 3 years In addition 15 outpatients were put on chlorpropamide during our clinical investigation

In July 1958 we were able to report on 136 patients studied to that time We have now enlarged this group to 200 patients who either had been on insulin or whose diabetes had not been satisfactorily controlled by dietary restriction alone even though they were hospitalized The above figure also includes those patients who had been treated with tolbutamide carbutamide or both TABLE 1 shows that 146 patients (more than two thirds of the diabetics who appeared suited to oral therapy) could be well controlled with chlorpropamide Of the 146 patients 63 had not been controlled by insulin the remaining 83 had been Of the latter 40 received insulin only on

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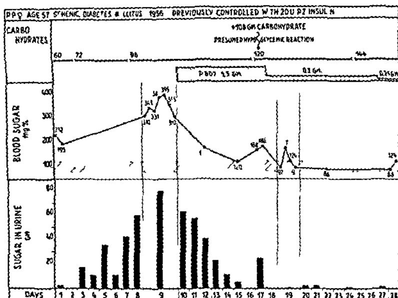


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Also evident is the fact that in patients taking insulin the possibility of success with chlorpropamide decreased as the need for insulin increased In the majority of cases requiring less than 40 U insulin daily chlorpropamide

could be substituted with success However, even in 21 diabetic patients requiring as much as 60 U of insulin daily, we succeeded in achieving good metabolic equilibrium with chlorpropamide Regarding duration of the disease and, to a lesser extent, duration of insulin medication quite favorable

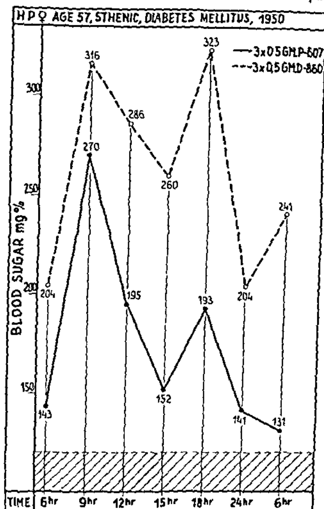


FIGURE 2

exceptions to the 10 and 5 year rule respectively could be obtained with chlorpropamide as was demonstrated in 26 diabetics in whom we replaced insulin completely although they had been receiving insulin for 10 to 20 years Recent diabetics even those of the asthenic type sometimes did well on chlorpropamide if they were well beyond 40 years of age

Regarding the responses in the two large diabetic groups the brittle and the stable, it may be stated that, in general the same criteria are applicable

with chlorpropamide as with the oral antidiabetic drugs previously used However as with other drugs, exceptions exist and because mixed forms result from the overlapping of the classifications brittle and stable the exceptions increase

Observations

The effect of chlorpropamide in reducing blood sugar and sugar in urine is greater than that of the two other oral antidiabetics now in use Hence

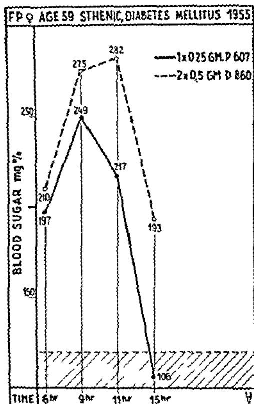


FIGURE 3

with a lower dosage of chlorpropamide (sometimes substantially lower especially during the initial treatment period) equally favorable therapeutic results can be achieved together with good equilibrium of metabolism (FIGURES 2 3 4 and 5) The initial daily dosage usually was 1 gm (two 500 mg doses) and in some cases 1 5 gm (three 500 mg doses) Maintenance dosage was usually 0 5 gm or 0 25 gm frequently given every other day In patients on relatively low quantities of carbohydrates (for example 80 to 120 gm daily) higher initial dosage can lead to sudden hypoglycemic episodes

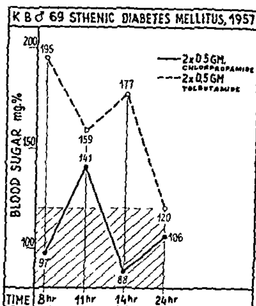


FIGURE 4

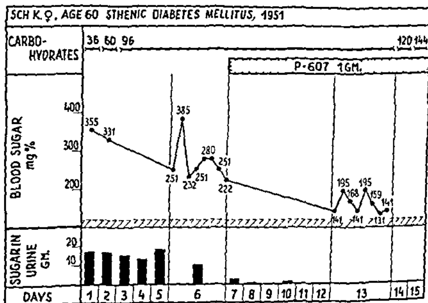


FIGURE 5 Example of good control with chlorpropamide Patient previously controlled with 20 U lente insulin is now controlled with two 0 5 gm doses carbutamide daily

especially if the treatment is preceded by a considerably restricted, although short term, diet (48 to 60 gm carbohydrate)

The following episode supports the theory (of the mechanism of action of the oral antidiabetic drugs) that these preparations promote or activate endogenously produced insulin The dextrose loading test carried out in a nondiabetic (FIGURE 6) who because of obesity was put on a restricted diet (especially with respect to carbohydrates) showed very good endogenous

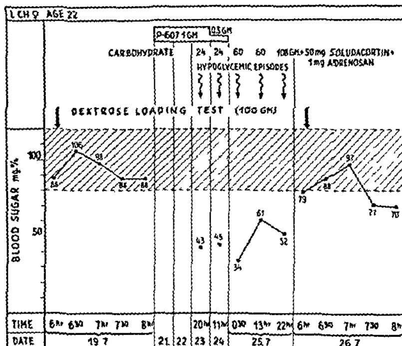


FIGURE 6 Promotion by chlorpropamide of endogenously produced insulin

insulin action After the administration of six 500 mg doses of chlorpropamide in a period of 3 days the patient suddenly experienced a serious convulsive hypoglycemic episode accompanied by unconsciousness and a decrease in the blood sugar level to 43 mg per cent During the next day the patient remained in a serious hypoglycemic condition blood sugar levels varying between highs of 34 and 61 mg per cent The hypoglycemia could be combated only by continuous glucose administration (260 gm) part being given intravenously We also administered injections of Soludacortin and adrenalin A repeat of the loading test done with 100 gm of dextrose and performed on the third day brought about blood sugar levels of 70 to 88 mg per cent, with one peak of 97 mg per cent

Because of this markedly greater hypoglycemic action it would be possible

to treat with smaller doses of chlorpropamide diabetics who require far more exogenous insulin, this would not generally be the case with tolbutamide This action, however, may be regarded as an advantage only if prolonged use and careful laboratory testing show that the chronic toxic effects of the drug or its degradation products have no deleterious effects, particularly on the liver, kidneys, and the hematopoietic systems The problem of toxicity in this regard is of special interest because we may assume that the ability of chlorpropamide to reduce blood sugar is due to its considerably delayed excretion and that the prolonged effectiveness of the lower dosages is due primarily to cumulative action

In view of the above evidence and its substantiation by clinical observation, we can state that the initial adjustment period for chlorpropamide is longer (as much as one week) and the duration of action more prolonged than with tolbutamide

Of 215 patients (including outpatients) untoward side reactions were observed more often at the beginning of the treatment and less frequently after therapy had been in progress for some time Gastrointestinal disturbances were noted in 10 7 per cent of cases, dermatological side reactions in 3 5 per cent and intolerance to alcohol in 11 6 per cent

Gastrointestinal disturbances occurred in twenty three cases and manifested themselves as loss of appetite (often to a great degree), malaise, and occasional nausea and vomiting Sometimes these disturbances were associated with other complaints in the upper abdomen (for example, pressure on the stomach gastric cramps and bloating) and, although in only a few cases, with diffuse abdominal colic and constipation Incidence of these side reactions lowered dramatically when the initial dosage or maintenance dosages were reduced substantially This, however, necessitated a prolonged initial period of adjustment Thus the disturbances proved to be dependent on

of the skin was noted in 4 The latter symptom never became so marked that it necessitated discontinuation of the preparation, and it subsided without requiring therapy In 2 cases of morbilliform exanthemas, we were able to continue medication under antiallergic protection Marked changes in the differential blood count of the white blood corpuscles (for example, the eosinophils) were not observed in association with the above skin reactions

As to the hematopoietic system, no toxic effects were noted except for occasional granulocyte decrease which, as proved by periodic follow ups, was only transitory No hepatic damage or adverse effect on the kidneys or the central nervous system became manifest during the time of observation, which was as long as five months

Flushing, subjectively often felt as "congestion" and a warmth of the face and neck, visible as a temporary redness sometimes extended to the upper thoracic area It was observed in 31 patients (regularly in 24) after consumption of even a small quantity of alcohol The above-mentioned symptoms generally occurred 10 to 15 min after taking a small quantity of alcohol,

regardless of the type of alcohol imbibed and were similar to an Antabuse alcohol reaction Of course these side reactions may be prevented by avoidance of alcohol

A few patients complained of giddiness and buzzing in the ears these symptoms however cannot be clearly designated as side reactions because their interpretation allows so many other possibilities to be taken into consideration

Conclusion

It may be stated that chlorpropamide has two advantages over the other antidiabetic sulfonylurea preparations first minimal dosages will produce an entirely satisfactory hypoglycemic effect that compares favorably with that of the other antidiabetic drugs previously used and second it is distinguished by an especially prolonged duration of hypoglycemic action so that in no case need it be given more often than once a day and it may be given as infrequently as every other day A higher incidence of dyspeptic disturbances must be looked upon as a disadvantage however these disturbances were observed principally during initiation of therapy when relatively high doses were being given

CHLORPROPAMIDE IN THE MANAGEMENT OF THE ADULT DIABETIC PATIENT*

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This article will describe our experiences with chlorpropamide in the clinical management of adult diabetic patients

Methods and Results

Our observations were made on 32 diabetic patients who received chlorpropamide for as long as 36 weeks (FIGURE 1) The study included 20 females and 12 males, with an age range of 33 to 74, 26 were in the range of 50 to 70

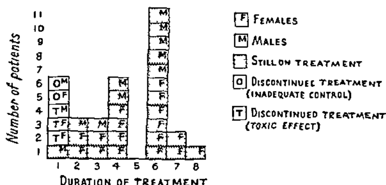


FIGURE 1 Duration of chlorpropamide therapy in 32 patients

(TABLE 1) Constant dietary regimen has been maintained, most of the patients having 2000 cal diets consisting of 250 gm carbohydrate, 90 gm protein, and 70 gm fat—divided into meals of approximately equal caloric value This basic diet plan has been changed in a few cases where body weight indicated need for adjustment Thirteen patients were observed initially in the hospital and 19 have been handled entirely as outpatients Diabetic control has been checked by frequent fasting and 2 hour postbreakfast plasma glucose analyses[1] and frequent determinations of 24 hour glycosuria [2]

A systematic study of possible toxicity has been carried out and has included frequent clinical observation of the patient There have been periodic determinations of body weight serum cholesterol, BSP retention, total and fractional serum proteins, cephalin cholesterol flocculation, serum alkaline phosphatase, icterus index, blood urea nitrogen, urine analysis, and platelet and complete blood counts There have been occasional prothrombin time determinations Serum diastase, transaminase, and bili

* The work on which this paper is based was supported by a grant from Chas Pfizer & Co, Brooklyn N Y, who supplied the chlorpropamide used in the study

rubin levels have been investigated in a few of the cases Needle biopsies of the liver al o have been obtained in a few cases

In general our earlier cases received initial doses of 1 5 to 1 0 gm The frequency of mild hypoglycemic reactions caused us to decrease the initial dosage to 0 5 gm or less by the time we had had experience with the first 15 patients (TABLE 2) Only 1 patient received an initial dose of more than 1 0 gm Chlorpropamide was discontinued in 2 patients because of inadequate control, both had had good control on insulin In one chlorpropamide was started with progressive decreases in insulin but the improvement was

TABLE 1
AGE DISTRIBUTION AND STATE OF CONTROL OF 32 PATIENTS WITH DIABETES MELLITUS

Age range	No of cases	Control on chlorpropamide	
		Adequate	Inadequate
30–40	1	1	0
40–50	4	4	0
50–60	11	10	1
60–70	15	14	1
70 80	1	0	1
Total	32	29	3

transient as demonstrated by increasing glycosuria and insulin requirements In the other case brought to the hospital in a state of insulin produced hypoglycemia an attempt made to control the patient with chlorpropamide alone was unsuccessful Chlorpropamide was discontinued in 3 cases because of possible toxicity In 2 others chlorpropamide was discontinued temporarily and resumed without further difficulty when it was decided that accompanying skin reactions were unrelated to the drug

Clinical Observations

In assessing control the same criteria were used as in judging effectiveness of tolbutamide in the therapy of adult diabetes mellitus[3,4] Adequate control was designated good or fair A patient was considered to have a good response if one of the following phenomena was observed a disappearance of the insulin requirement in the insulin treated patients a disappearance of glycosuria with an associated decline in plasma glucose levels in the noninsulin treated glycosuric patients or a marked fall in the fasting and postprandial plasma gluco e levels in the aglycosuric patients when compared to similar levels without chlorpropamide These biochemical aspects of control were considered together with the following clinical objectives clinical well being at all times (that is freedom from any symptoms) achievement and maintenance of ideal body weight, absence of ketosis and hypoglycemic reactions A given patient was considered to have a fair

TABLE 2
SUMMARY OF 32 ADULT DIABETIC PATIENTS TREATED WITH CHLORPROPAMIDE

Case No	Sex	Age	Duration: Diabetes (years)	Duration: Insulin treatment	Duration: Tolbutamide treatment	Complications	Date of initial treatment	Date treatment discontinued	Initial dose (gm)	Maintenance dose (gm)	Response*
1	F	66	3	—	—	Paget's disease cataracts	11 30 57		1 0	0 1	G
2	F	57	0	17 days	—		1 58		0 5	0 1	G
3	F	51	10	2½ years	—	Retinopathy mild	1 16 58		1 0	0 5	F
4	F	33	0	0	0	Obesity	1 20 58		1 5	0 5	F
5	M	61	20	10 years	2 weeks	ASCVD retinopathy	2 27 58	3 20 58	1 0	1 0	G
6	M	70	19	—	2 months	ASCVD	1 23 58		1 0	0 25	G
7	F	46	1	0	6 months		1 24 58		1 0	0 5	G
8	M	66	20	5 years	—	ASCVD arteriosclerosis obliterans	1 28 58		1 0	0 5	F
9	M	52	3	3 years	—	ASCVD retinopathy	2 6 58		1 0	0 25	G
10	M	68	18	20 years	6 months	ASCVD	2/6/58		1 0	0 5	G
11	M	53	14	—	—	Retinopathy neuropathy	2/9 58		1 0	0 25	G
12	F	65	1	1 year	—	ASCVD cataracts	2/13/58		1 0	0 2	G
13	F	63	20	20 years	—	ASCVD	2/13/58		1 0	0 5	F
14	F	45	—	—	—	Gallbladder disease with stones	2 24/58		1 0	0 1	G
15	F	62	22	16 years	—	ASCVD hypertension	2/20/58		0 5	0 5	F
16	M	68	5	5 years	—		4/3/58		0 5	0 1	G
17	F	46	—	—	—	Obesity hypertension	4/9/58	7/1/58	0 25	0 1	G
18	F	46	3	—	—	Retinopathy	4/10/58		0 5	0 1	G
19	M	49	3	3 years	10 months		4/17/58		0 5	0 5	F
20	M	74	2	—	—	ASCVD	4/17/58		0 5	0 5	F
21	F	59	2	—	—		4/17/58		0 1	0 3	G
22	M	58	—	—	—	ASCVD	4/19/58		0 25	0 25	F

TABLE 2 (*Continued*)

Case No	Sex	Age	Duration			Complications	Date of initial treatment	Date treatment discontinued	Initial dose (gm)	Maintenance dose (gm)	Response*
			Diabetes (years)	Insulin treatment	Tolbutamide treatment						
23	F	63			—	Obesity	4 30 58		0 25	0 2	G
24	M	64	4		—	ASCVD	5 12 58		0 1	0 1	G
25	F	59	8		—	ASCVD	7 3 58	7 17 58	0 25	0 25	G
26	M	64			—	ASCVD coronary disease	5 29 58		0 2	0 2	G
27	F	55					6 12 58		0 25	0 5	G
28	F	58	5		1 year		6 17 58		0 5	0 5	G
29	M	59	26	26 years		Cataracts osteoarthritis	6 20 58	7 11 58	0 5	0 5	O
30	M	69	10	10 years			7 4 58		0 5	0 5	G
31	F	45	7	1 year		Hypertension osteoarthritis	7 11 58	7 31 58	0 5	0 5	G
32	F	67	22	22 years		ASCVD neuropathy	7 14 58	7 23 58	0 5	0 5	O

* Key G good F fair O inadequate

TABLE 3
CONTROL ACHIEVED WITH CHLORPROPAMIDE THERAPY

Patients	Adequate		Inadequate	Total
	Good	Fair	Poor	
Number	22*	8	2	32
Percentage of total	68 7	25 0	6 3	100

* Includes three patients who showed good results for 2 to 3 weeks but in whom treatment was discontinued because of toxicity

response when the above criteria were partially fulfilled TABLE 3 summarizes our experience Thirty of our patients showed adequate control 22 showed very good response, and 8 showed a fair or partial response Two patients showed no response both were diabetics of long standing (more than 20 years), with substantial insulin requirements

This investigation has little statistical significance However, observations in a few cases raised interesting questions Case 11, M P, is a 53 year old male who had had mild diabetes for 14 years and who had had no previous therapy other than avoidance of "sweets" (FIGURE 2) Outpatient observations had established fasting plasma glucose levels at approximately 275 mg per cent and 2 hour postbreakfast levels of more than 350 mg per cent His response to 1 0 gm of chlorpropamide was immediate, the dose was

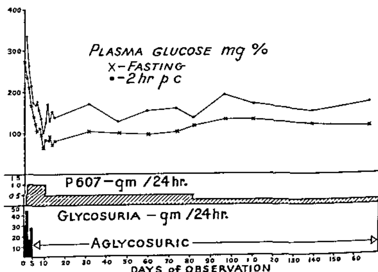


FIGURE 2 Case 11 M P male, 53 years old Duration of diabetes 14 years No previous treatment

reduced to 0 5 gm on the tenth day because the plasma glucose level had fallen to 66 mg per cent fasting and 105 mg per cent 2 hours after breakfast On this dosage he was observed as an outpatient plasma glucose levels remained in the range of 100 to 110 mg per cent fasting and 130 to 160 mg per cent 2 hours after breakfast Further reduction in chlorpropamide to 0 25 gm once daily before breakfast has resulted in slight increase in the

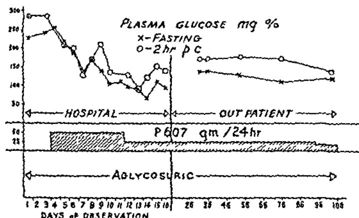


FIGURE 3 Case 18 I R female 46 years old diabetic 3 years Previous treatment diet

Case 18, I R a 46 year old female with diabetes of 3 years duration and treated by diet alone was aglycosuric throughout our observations (FIGURE 3) Her response in the plasma glucose levels to the initial dose of 0 5 gm chlorpropamide was dramatic and compelled reduction to 0 25 gm on the ninth day There has been progressive weight loss from 173 to 163 lb and the patient is well maintained currently on 0 2 gm chlorpropamide daily

Case 26, A G a 64 year old male with newly discovered diabetes was handled entirely as an outpatient His pretreatment fasting and 2 hour

satisfactory response

The postprandial plasma glucose level as an indicator of therapeutic effectiveness impressed us in evaluating tolbutamide therapy [3,4] The three cases presented illustrate the same phenomena and the longer term observations support our impression that the prompt and substantial fall in the postprandial plasma glucose level bespeaks a good total response

The information that approximately five days were required on a given

dosage of chlorpropamide to achieve a blood level plateau, and that the levels fell slowly in the two or three days following cessation of the drug immediately suggested the possibility that we might observe in some of our cases a ' chlorpropamide hang over" effect in the plasma glucose levels[5] We believe we have observed this in one case

Case 31, S T is a 65 year old white female, diabetic for 7 years who had been managed for the most part with diet alone, but who had also taken as much as 15 U of insulin for short periods (FIGURE 5) Initially observed in the hospital she seemed resistant to 0 5 gm chlorpropamide given as a single daily dose before breakfast After 6 days of this therapy, the fasting plasma glucose level was 189 mg per cent and the 2 hour postbreakfast level was

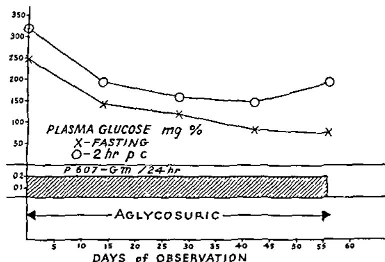


FIGURE 4 Case 26 A G male 64 years old new diabetic

235 mg per cent She received placebo therapy the next 2 days, on the third day, plasma glucose levels were 145 mg per cent and 246 mg per cent respectively The relatively lower fasting plasma glucose level and the virtually unchanged 2 hour postprandial level after 2 days of placebo therapy suggested the possibility of a ' hang over effect The more favorable *response when the drug was resumed* remains unexplained There have been no correlation studies between serum levels of chlorpropamide and plasma glucose levels Observations in this case have led us to institute such studies If the observation is validated chlorpropamide may have a significant potential for successful intermittent therapy

Observations Concerning Toxicity

With respect to toxicity, the last case that of S T showed a cephalin cholesterol flocculatio ... after instituting chlorpropamide thera[...] transaminase levels ,

results In this case we were fortunate to have needle biopsies of the liver before starting chlorpropamide therapy, and these were repeated as soon as we had confirmed the three-plus cephalin cholesterol flocculation reaction No evidence of necrosis or any other change was detected with hematoxylin and eosin stain One other case presented the same cephalin cholesterol flocculation abnormality with no other abnormality in the liver profile Needle biopsies of the liver were not obtained In both cases the drug was stopped immediately

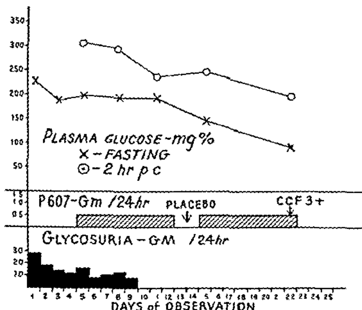


FIGURE 5 Case 31 S T female 65 years old diabetic 7 years

In one other case the drug was stopped because the patient complained of nausea and a constant disagreeable metallic taste This patient had done well previously on tolbutamide and at his insistence was returned to this therapy

Summary

Thirty two adult diabetic patients receiving chlorpropamide have been observed closely for periods as long as 36 weeks Thirty of the patients showed adequate response 22 showed a very good response and 8 a fair response Two of those showing a good response developed positive cephalin cholesterol flocculation reactions after about 3 weeks of therapy and the drug was discontinued Two showed no response both of these had had diabetes for more than 20 years and had had substantial insulin requirements No patients with juvenile type of diabetes were included in these studies

During observations it was noted in one instance that there was an appar

ent hang-over' effect on blood sugars from the chlorpropamide given forty-eight hours previously. The possibility that intermittent therapy is feasible is suggested and is being studied. Normal needle biopsy liver tissues before therapy and at the time one patient developed a positive cephalin cholesterol flocculation reaction show a dissociation between biopsy results and liver function tests. With available tests, hepatotoxicity is extremely difficult to assess.

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COMPARATIVE HYPOGLYCEMIC EFFECTS OF CHLORPROPAMIDE, TOLBUTAMIDE AND FURFURYLUREA*

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This study of the relative potency of the several hypoglycemic agents in altering the blood sugar and serum electrolytes in diabetes mellitus also permits some speculation on the possible sites of action in individual patients

Materials and Methods

Observations were made on adult as well as juvenile diabetic patients Age at the onset of diabetes and at the time of these studies the sex of the patient and the dosage and type of insulin taken if any are recorded in TABLE 1

Chlorpropamide tolbutamide or furfurylurea † 3 0 or 1 0 gm was administered after an overnight fast to 9 juvenile and 17 adult diabetics The venous blood sugar and the levels of inorganic phosphorus CO_2 potassium sodium, and chloride in serum from venous blood were measured at hourly intervals for 5 hours in control studies made both prior to and after the ingestion of the hypoglycemic agent One week or more elapsed between the administration of chlorpropamide tolbutamide or furfurylurea All studies were conducted on an outpatient basis after maintaining the subjects on rapid-acting insulin for 2 or more days

Results

Control studies From the control observations (FIGURE 1) it may be seen that in most patients the blood sugar levels remained relatively close to the zero value or declined slightly In a few patients a distinct spontaneous rise or a definite fall, that is, 40 mg per cent or more was recorded (FIGURE 1) These limits were selected because such changes were rare in the control periods there were but 2 such patients in each category These were not confined to either the juvenile or adult group and were unrelated to either the type of insulin employed prior to these studies or to the usual daily dosage of insulin

Chlorpropamide effects (3 0 gm dosage) Chlorpropamide decreased the fasting blood sugar levels 40 mg per cent or more in 14 of the patients including 2 of the juvenile diabetics In 2 others 1 adult and 1 juvenile type diabetic the blood sugar rose by 40 mg per cent or more Decreases were

* The study reported in this paper is part of the Pittsburgh Plan for Heart Disease Control and was aided by grants from the John A Hartford Foundation Inc New York N Y the Public Health Service Bethesda Md and Chas Pfizer & Co Inc Brooklyn N Y

† The structural formula of *furfurylurea is 1 p-tolylsulfonyl 3 furfurylurea* This compound was supplied by The National Drug Company Philadelphia Pa

recorded most consistently in patients in whom the starting blood sugar was between 120 and 260 mg per cent (FIGURE 2)

Comparisons of chlorpropamide, tolbutamide, and furfurylurea effects at 3 0 and 1 0 gm dosages Seven of the patients given 3 0 gm of chlorprop-

TABLE 1

Patient	Sex	Age, years	Age at onset of diabetes, years	Usual insulin dosage, U
			Juveniles	
W C	M	13	10	25 (NPH)
N G	M	13	12½	3 (NPH)
R L	M	33	10	24 (Protamine Zinc I) 54 (Regular)
D M	M	14	13	No insulin
E M	F	16	12	46 (Lente)
S M	M	10	7½	30 (Crystalline)
L P	M	16	16	12 (NPH)
C R	F	21	2	65 (Protamine Zinc)
D S	M	16	13	40 (Protamine Zinc) 60 (Regular)
			Adults	
G C	F	32	25	18 (NPH)
W E	M	32	20	50 (NPH)
J E	F	24	1	42 (Protamine Zinc) 22 (Regular)
A H	F	60	60	No insulin
P K	F	76	70	15 (Protamine Zinc)
A L	F	43	27	44 (NPH)
A M	F	63	63	No insulin
M M	F	49	40	25 (Lente)
B P	F	56	46	30 (NPH)
J S	F	56	56	No insulin
R N	F	54	46	No insulin
H F	F	55	47	No insulin
L V	F	58	48	No insulin
C S	F	56	55	No insulin
J A	F	56	56	No insulin
L R	M	61	57	No insulin
A B	F	56	49	No insulin

amide were subsequently tested with similar amounts of tolbutamide and of furfurylurea, and then with 1 0 gm of chlorpropamide and of furfurylurea The blood sugar changes obtained are shown in FIGURE 3 TABLE 2 is based ... greater or less than minus 40 mg per cent together

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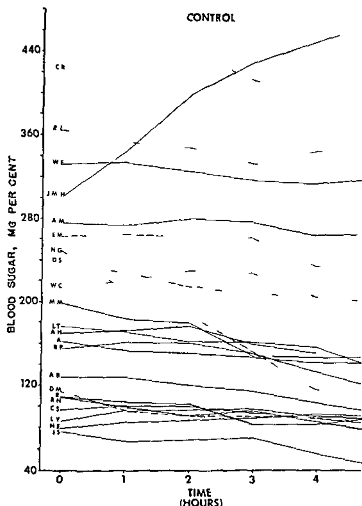


FIGURE 1 Serum blood sugar levels in series of adult and juvenile type diabetic patients. Dotted lines identify patients whose diabetes began during childhood. If a decrease in blood sugar of 40 mg per cent or more is taken to be significant, then such a decrement occurred spontaneously in only 2 patients.

producing a decrease of 40 mg per cent or more in 5. Tolbutamide in the same dosage produced similar effects in only 2 of the 7.

Again it should be noted that in general the responders by the criterion employed—a fall of 40 mg per cent or more—were patients with starting blood sugars of 120 to 260 mg per cent and that a greater proportion of values below 120 mg per cent was present during the tolbutamide trials.

It should be noted further that of the agents and the dosages tested only

furfurylurea lowered the blood sugar 40 mg per cent or more in 3 of 5 patients with levels of 120 mg per cent or less at zero time

Finally reducing the dosage of chlorpropamide from 3 gm to 1 gm produced a decrease of blood sugar of 40 mg per cent or more in only 1 of the

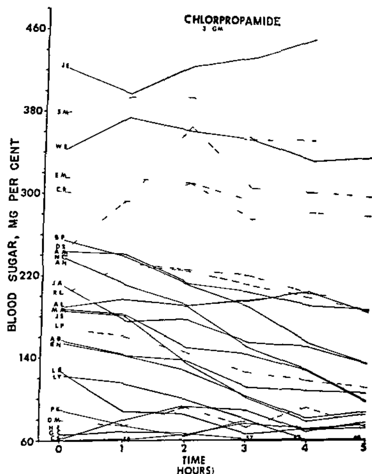
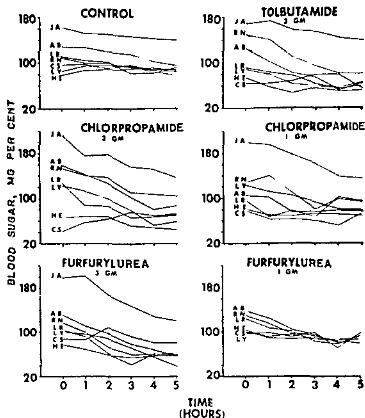


FIGURE 2 Hypoglycemic effects of chlorpropamide The administration of 3 0 gm of chlorpropamide at minus 45 min produced decreases in blood sugar of 40 mg per cent or more in 12 of the patients including 2 juvenile diabetics

patients In 1 other R N a change of this magnitude was partially cancelled by a subsequent rise This contrasts with the 3 0 gm studies in which all of the patients for whom such a comparison is available developed a distinct relative or absolute hypoglycemia Furfurylurea reduced to the dosage of 1 0 gm , decreased the responses to none of 6 in contrast to 5 of 7 with 3 0 gm

Serum inorganic phosphorus changes in relation to blood sugar alterations In FIGURE 4 the changes in blood sugar at 1 2 3, 4 and 5 hours in relation to



e starting blood sugar less than 120 mg per cent

ie fasting levels have been plotted against the increment or decrement in … If decrease of

rops in blood sugar in still other patients the blood sugar actually increased

Discussion

Although the data presented indicate that not all patients are equally ponsive to chlorpropamide or other oral insulin substitutes, decreases in

blood sugar can occur with chlorpropamide in both juvenile and adult type diabetes Insofar as relative efficacy in this regard is concerned chlorpropamide and furfurylurea appeared to be more effective than tolbutamide in this small group of patients It is noteworthy, too, that none of the other agents tested produced responses in patients unaffected by chlorpropamide (TABLE 2) Finally, although chlorpropamide and tolbutamide each appeared to be more effective when the zero value lay between 120 and 260 mg per cent furfurylurea alone acted at levels below 120 mg per cent

It is well established that serum inorganic phosphorus decreases quite regularly with the acceleration of carbohydrate metabolism in nondiabetic

Table 2

Hypoglycemic Responses in the Same Adult Diabetic Patients to Various Oral Insulin Substitutes

Patient	Control	Chlorpropamide 3 0 gm	Chlorpropamide 1 0 gm	Tolbutamide 3 0 gm	Furfurylurea 3 0 gm	Furfurylurea 1 0 gm
J A	–	+	+	–	+	
A B	–	+	–*	+	+	–
R N	–*	+	–	+	+*	–
L R	–*	+	–*	–*	+*	–
L Y	–*	+	+	–*	+*	–*
H E	*	–*	–*	–*	–*	–*
C S	*	–*	–*	–*	–*	–*
Blood sugar more than –40 mg per cent	0 7	5 7	2 7	2 7	5/7	0 6

* Zero blood sugar less than 120 mg per cent

patients by glucose insulin or glucose and insulin in combination and upon insulin injection in those diabetic patients previously shown to be unable to dispose of glucose loads at a normal rate and to develop a significant drop in inorganic phosphorus [1] This decrease in serum inorganic phosphorus can be taken to reflect disposal of glucose by anaerobic glycolysis and formation of high energy phosphorous compounds, since deposition of liver glycogen does not segregate phosphate [2,3]

It may be assumed that diabetic patients maintained on diet alone possessed some ability to elaborate insulin It would then appear, from those cases in which no significant decrease was observed in the serum inorganic phosphorus concomitant with the decrease in blood sugar, that chlorpropamide and the other agents tested had not increased anaerobic glycolysis (FIGURE 4) The observed decreases in blood sugar without concomitant hypophosphatemia are to be attributed therefore to decreased glycogenolysis and gluconeogenesis or increased deposition of glucose as liver glycogen, or both, a process that, unlike anaerobic glycolysis does not preempt inorganic phosphorus from the serum In patients in whom such hypoglycemic changes occurred with a concomitant drop in inorganic phosphorus, anaerobic glycolysis

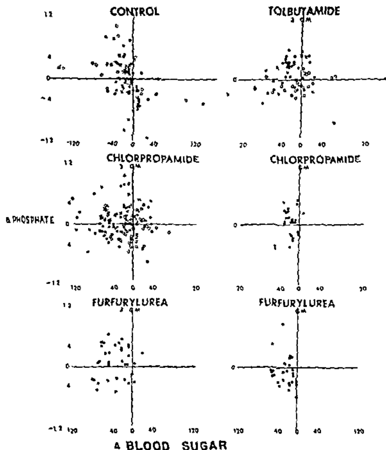


FIGURE 4 Correlation of serum inorganic phosphorus and blood sugar changes in [illegible] s identity adult [illegible] on insulin which [illegible] and open circles [illegible] It is apparent that on many occasions the blood sugar decreased without concomitant significant fall in serum inorganic [illegible] 0.1 mg per cent or more. These decreases may be [illegible] enous insulin *or from accentuation or prolongation of the effect of such endogen us insulin*

presumably was accelerated either by discharging insulin, by accentuating its action or survival or by increasing the permeability of cells to glucose

These comparisons are not offered, of course with any assurance that the drugs tested would necessarily prove effective in reducing the insulin requirement or in regulating the diabetes in these patients Also, it is possible that

in keeping with the experience of others with agents other than chlorpropamide, some of the patients who did not show a positive response to a particular drug initially might do so after several weeks of treatment [6]

Summary and Conclusions

It is possible that responses to single dosages of oral hypoglycemic agents may differ from those obtained after repeated or prolonged therapy The following statements therefore apply only to the single dosage experiments herein described

Certain diabetics did not respond to single dosage of chlorpropamide tolbutamide or furfurylurea with a decrease in blood sugar

Chlorpropamide administered in 3 gm doses at minus 45 min produced during the next 5 hours a decrease of 40 mg per cent or more in the zero blood sugar level of most of the adult diabetics and in 2 of 7 juvenile diabetics or patients whose diabetes began during childhood During control observations comparable decreases occurred only occasionally

Chlorpropamide and furfurylurea tested in equal dosages in the same patients produced greater response than did tolbutamide

Decreases with any of the agents tested were noted most consistently in patients in whom the zero blood sugar level lay between approximately 120 and 260 mg per cent

Furfurylurea may differ from the other agents in possessing a greater ability to lower blood sugar levels when the starting values are less than 120 mg per cent

The absence of significant decreases in the serum inorganic phosphorus in patients in whom the blood sugar fell significantly would suggest that in general the hypoglycemic responses in this group of diabetics are attributable to decreased gluconeogenesis and glycogenolysis or to segregation of glucose in the liver as glycogen a process that unlike anaerobic glycolysis does not lower serum inorganic phosphorus In those in whom both the blood sugar and the serum inorganic phosphorus fell, an increase in anaerobic glycolysis can be postulated—this as a result of a greater output survival or effectiveness of endogenous insulin or as a consequence of an increase in the permeability of cells to glucose

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CHLORPROPAMIDE IN SINGLE DOSAGE AND THE RESPONSES TO INSULIN OR GLUCOSE IN DIABETICS THE ABSENCE OF EVIDENCE OF INCREASED GLUCOSE UTILIZATION*

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In responsive patients with diabetes mellitus, chlorpropamide in single dosage lowers blood sugar levels, often without a concomitant decrease in serum inorganic phosphorus [1] This suggests that, in patients who develop relative or absolute hypoglycemia after a single dose without significant hypophosphatemia, the chief or, perhaps sole action of this agent is segregation of glucose as glycogen (a process not associated with deposition of phosphate) or suppression of gluconeogenesis or glycogenolysis To examine further the possibility that there is a simultaneous increase in glucose utilization, glucose tolerance tests and insulin tolerance tests with and without

TABLE 1

Patient	Sex	Age, years	Ages at onset of diabetes years	Usual insulin dosage, U
			Juveniles	
N G	M	13	12½	3 (NPH)
R L	M	33	10	24 (Protamine Zinc I)
			14	54 (Regular)
D M	M	14	13	No insulin
F M	F	16	12	46 (Lente)
S M	M	10	7½	30 (Crystalline)
L P	M	16	16	12 (NPH)
C R	F	21	2	65 (Protamine Zinc)
D S	M	16	13	40 (Protamine Zinc)
				60 (Regular)
			Adults	
G C	F	32	25	18 (NPH)
W E	M	32	20	50 (NPH)
A L	F	43	27	44 (NPH)
M M	F	49	40	25 (Lente)
B P	F	56	46	30 (NPH)
J McH	M	37	21	28 (Regular)
J S	F	56	56	No insulin

* The study reported in this paper is part of the Pittsburgh Plan for Heart Disease Control, and was aided by grants from the John A Hartford Foundation Inc, New York, N Y, the Public Health Service, Bethesda Md and Chas Pfizer & Co, [illegible] Brooklyn N Y

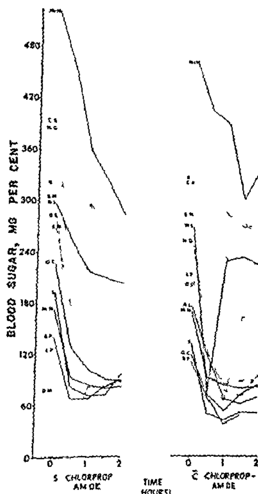


FIGURE 1. Blood sugar changes during insulin tolerance test in diabetics with and *without 3.0 gm. of chlorpropamide at minus 45 min.* Comparison of the changes at 0, ½, 1, 1½, and 2 hours in the 2 groups shows no accentuation of hypoglycemia after chlorpropamide (TABLE 2). Broken lines identify juvenile diabetics.

chlorpropamide administration in single dosage were undertaken, and the changes in serum inorganic phosphorus and other electrolytes were measured.

Materials and Methods

The clinical histories of the diabetic patients studied (some with juvenile diabetes mellitus and others with diabetes that began during the adult years) are summarized in TABLE 1. In most of the glucose tolerance tests 0.5 gm. of glucose per kilogram of body weight was administered intravenously at zero time as a 50 per cent solution in water. In a few patients 1.75 gm. of glucose per kilogram of body weight was used *orally*. In the

insulin tolerance tests 0.1 U. of commercial rapid acting crystalline insulin (per kilogram of body weight), diluted 1:9 with normal saline for ease of measurement, was injected intravenously at zero time. Chlorpropamide (3.0 gm.) was then given 45 min. before the start of the second glucose or insulin tolerance test. Blood sugar and serum electrolytes were measured just prior to and at intervals after the administration of the sugar or the insulin.

TABLE 2

STATISTICAL EVALUATION OF BLOOD SUGAR AND SERUM INORGANIC PHOSPHORUS CHANGES DURING INSULIN TOLERANCE TESTS IN DIABETIC SUBJECTS

Period	Hours	No. of patients	Mean	±S.D.
		Blood sugar		
Control	½	12	−84.9*	35.9
Chlorpropamide	½	12	−5[illegible]	39.8
Control	1	12	110.8	43.6
Chlorpropamide	1	12	−87.8	42.6
Control	1½	12	−119.8	46.0
Chlorpropamide	1½	12	−8[illegible].7	4[illegible].9
Control	2	11	−126.8	51.8
Chlorpropamide	2	11	−88.8	42.6
		Serum inorganic phosphorus		
Control	½	10	−0.590	0.445
Chlorpropamide	½	10	−0.451	0.236
Control	1	10	−0.649	0.388
Chlorpropamide	1	10	−0.440	0.391
Control	1½	11	−0.509	0.423
Chlorpropamide	1½	11	−0.397	0.434
Control	2		0.570	0.322
Chlorpropamide	2		−0.294	0.416

* Difference is statistically significant

Results

Insulin tolerance tests. FIGURE 1 summarizes the blood sugar changes in the insulin tolerance tests with and without the prior administration of 3.0 gm. chlorpropamide at minus 45 min. Patients whose diabetes began during childhood are indicated by broken lines. The solid lines identify [illegible] usually required insulin in the dosages

was equally true of the juvenile or formerly juvenile diabetics

adult diabetics In fact, the blood sugar decrease at one half hour was somewhat less after chlorpropamide (TABLE 2) It is probable that the week-to-week fluctuations in the starting blood sugar levels obscured the decrement often produced in fasting levels by chlorpropamide Nevertheless, the fact

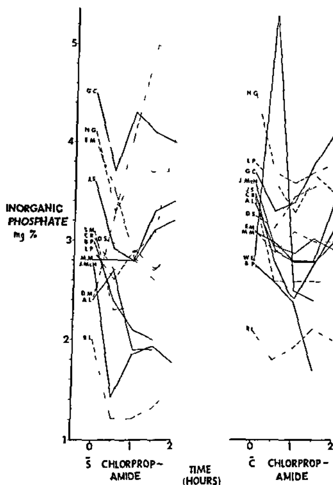


FIGURE 2 Serum inorganic phosphorus changes during insulin tolerance test in diabetics with and without 3 0 gm of chlorpropamide As in the blood sugar changes there is no evidence that the serum inorganic phosphorus responses are regularly affected by chlorpropamide (TABLE 2)

that insulin preceded by chlorpropamide failed to produce a decrement in blood sugar greater than that seen with insulin alone argues against an augmentation of insulin action by this oral insulin substitute when it is given in single dosage It also eliminates the possibility that a single dosage of chlorpropamide enhanced endogenous insulin output in these particular

patients, it provides no information of course on the effects of repeated doses or prolonged therapy

Chlorpropamide did not increase the magnitude of the hypophosphatemia produced by insulin alone (FIGURE 2 TABLE 2) Since increased utilization of glucose by means of anaerobic glycolysis increases the drop in serum inorganic phosphorus, the apparent absence of such a change in these experiments can be cited as further evidence against a release of endogenous insulin

TABLE 3
STATISTICAL EVALUATION OF BLOOD SUGAR AND SERUM INORGANIC PHOSPHORUS CHANGES DURING I.V. GLUCOSE TOLERANCE TESTS IN DIABETIC SUBJECTS

Period	Hours	No. of patients	Mean	S.D.
		Blood Sugar		
Control	½	8	123	8.3
Chlorpropamide	½	8	88.1	[illegible] 4
Control	1	8	104.0	51.4
Chlorpropamide	1	8	65.[illegible]	60.[illegible]
Control	1½	8	[illegible]	55.[illegible]
Chlorpropamide	1½	8		68.2
Control	2	8	52.1	58.9
Chlorpropamide	2	8	38	60
		Serum inorganic phosphorus		
Control	½	8	0.228	0.206
Chlorpropamide	½	8	0.064*	0.190
Control	1		0.240	0.253
Chlorpropamide	1	7	−0.011*	0.158
Control	1½	8	−0.262	0.263
Chlorpropamide	1½	8	−0.184	0.220
Control	2	8	−0.322	0.224
Chlorpropamide	2	8	0.136*	0.281

* Differences are statistically significant

or prolongation or potentiation of the action of exogenous insulin by chlorpropamide in single dosage

Intravenous glucose tolerance tests The administration of chlorpropamide 45 min before the intravenous glucose produced a decrease in the increment in the blood sugar at the half hour point (TABLE 3)

The administration of chlorpropamide decreased the usual hypophosphatemic response to glucose loads The comparisons of the actual changes in blood sugar and serum inorganic phosphorus recorded during the glucose tolerance tests are summarized in TABLE 3 The differences in the blood sugar at the half hour, and in the serum inorganic phosphorus at the half, one-, and two hour points are statistically significant

It would appear that under the conditions of these experiments chlorpropamide did reduce the blood sugar response at the half hour point, but it

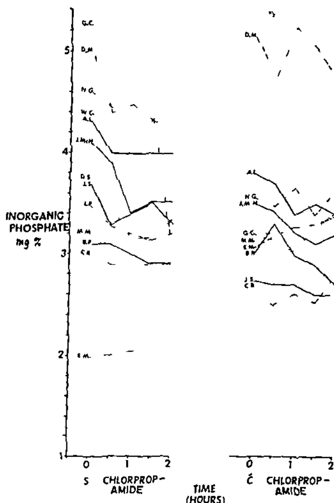


FIGURE 3 Blood sugar changes during glucose tolerance tests in diabetics with and without 3 0 gm of chlorpropamide at minus 45 min Solid lines identify adult diabetics and broken lines juvenile diabetics who received 0 5 gm of glucose intravenously at zero time Dotted lines represent patients who received 1 75 gm of glucose per kilogram of body weight by mouth at zero time In the intravenous glucose tolerance tests chlorpropamide decreased the degree to which the blood sugar rose at the half hour point (TABLE 3)

propamide in single dosage

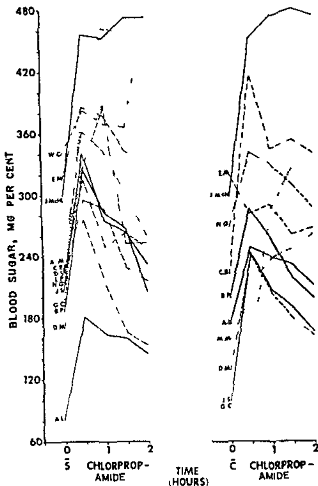


FIGURE 4 Serum inorgan phosphorus changes dur ng glucose toleran te t w th an l without 3 0 gm of chlorpropam le Symbols are as len fed in FIGURE 3 Th ser m inorganic phosphorus response to intravenous glucose was less al er chlorj rop am le (no e the ½ 1 and 2 hour points in TABLE 3)

the serum total CO_2 content [2] In diabetics not receiving insul n the admin istration of gluco e intravenously tends to rai e the serum potassium and lower the serum total CO_2 content [2] The e responses were not altered by chlorpropamide administration

Discussion

In these studies in keeping with findings on analysis of ti sues [3 6] it has been assumed that deposition of liver glycogen pre empts potassium but not

inorganic phosphorus The concomitants of muscle and other tissue glycogen formation are not known If it is true that glycogen deposition does not segregate inorganic phosphorus then the decrease in serum inorganic phosphorus reflects anaerobic glycolysis in cells and indicates either the formation of glucose phosphates or high energy phosphorus compounds or some other metabolic process that pre-empts this ion

In the studies herein described there was no evidence that the hypophosphatemic action of insulin or of glucose was enhanced by chlorpropamide in single dosage This finding argues against either a direct action of chlorpropamide in single dosage on anaerobic glycolysis or an effect upon exogenous or endogenous insulin The latter point is perhaps debatable since no direct information is available on the endogenous supplies of this hormone in our patients However it should be pointed out that in those of our subjects whose diabetes was controlled without insulin or with insulin in tiny amounts a significant residual capacity to produce insulin can be postulated However even these failed to show a greater drop and therefore no evidence can be brought forth in support of chlorpropamide action on the beta cells or on insulin secretion when the drug was administered in single dosage

Published studies designed to clarify the mechanism or mechanisms of action of the earlier oral hypoglycemic agents have at times led to diametrically opposed conclusions These and other discrepancies are undoubtedly attributable to variables such as species differences dosage and schedule variations and the characteristics of individual diabetics Confining ourselves to measuring the responses to a single and often large dosage of an oral hypoglycemic agent of the sulfonylurea type administered orally 45 min before the test procedure at intervals of 1 week or more, we have found that the hypoglycemic responses observed under these circumstances are frequently unaccompanied by a decrease in serum inorganic phosphorus This argues against a significant component of insulin discharge insulin potentiation insulin survival or direct action on cells in those instances of hypoglycemia since certainly at least the first three of these present in sufficient degree would be expected to lower the serum inorganic phosphorus Although the indirect nature of the evidence offered must be admitted the possibility is raised that the oral hypoglycemia agents of the sulfonylurea type elicit at least a 2 phase response Initially they act to suppress glycogenolysis and gluconeogenesis and segregate glucose as glycogen Endogenous insulin released in amounts too small to affect serum inorganic phosphorus could play a role in this, if such an action of insulin is accepted With continued therapy the beta cells are stimulated to elaborate insulin in quantities sufficient to produce the expected changes in serum inorganic phosphorus Such a sequence of events would account for some of the discrepancies in published reports of the effects or lack of effects of sulfonylureas on serum inorganic phosphorus or on other evidences of endogenous insulin production

Summary and Conclusions

In view of the fact that deposition of hepatic glycogen does not segregate phosphate in the liver the decrease in serum inorganic phosphorus which in

nondiabetics, invariably accompanies acceleration of carbohydrate metabolism by exogenous insulin or by glucose loads, can be taken as an index of enhanced disposal of glucose by anaerobic glycolysis

The failure to observe any increment in the hypophosphatemia characteristic of insulin or of glucose tolerance tests in these particular diabetic patients following chlorpropamide suggests that chlorpropamide in single dosage at least, did not accelerate anaerobic glycolysis either directly, by evoking endogenous insulin secretion, or by accentuating the action or survival of either exogenous or endogenous insulin Of course, this does not exclude such a response in other diabetics nor the possibility that repeated or prolonged therapy might provide evidence of an effect of chlorpropamide on anaerobic glycolysis unrelated or related to insulin

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CORTISONE ENHANCEMENT OF PERIPHERAL UTILIZATION OF GLUCOSE AND THE EFFECTS OF CHLORPROPAMIDE*

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Available evidence indicates that the hyperglycemia associated with excesses of ACTH and adrenocortical type steroids, which is largely attributable to increased gluconeogenesis, may be based in part on a blockade of insulin action or glucose utilization in the tissue.[1-5] An extensive series of

TABLE 1
FASTING HYPOPHOSPHATEMIA PRODUCED BY THE ADMINISTRATION OF CORTISONE 45 MINUTES BEFORE TEST

Sex	Cortisone mg	Blood sugar, mg %			Serum inorganic phosphorus*, mg %		
		Mean	±S.D.	No. tested	Mean	±S.D.	No. tested
		Healthy adults (21 to 60 years)					
M	0	84.3	11.1	64	3.43	0.54	67]
M	200	81.5	11.3	60	3.23	0.56	61]
F	0	80.7	11.6	71	3.58	0.55	70]
F	200	81.9	12.4	64	3.37	0.50	65]
		Relatives of juvenile diabetics (21 to 60 years)					
M	0	91.5	13.2	69	3.32	0.59	60]
M	200	87.9	10.9	26	2.97	0.49	26]
F	0	85.0	11.6	69	3.41	0.56	67]
F	200	84.5	9.6	27	3.12	0.46	27]

* Differences between the bracketed groups are statistically significant.

glucose tolerance tests (with serum inorganic phosphorus measurements) conducted in this laboratory on relatives of juvenile diabetics, on apparently healthy persons, on healthy dental students, and on adult diabetics suggests that under the conditions of our experiments cortisone probably enhances the net utilization of glucose by anaerobic glycolysis as a result of increased gluconeogenesis, by stimulation or enhancement of insulin action, or by facilitating the entry of glucose (alone or in combination) into cells.

* The study reported in this paper is part of the Pittsburgh Plan for Heart Disease Control, and was aided by grants from the John A. Hartford Foundation, Inc., New York, N. Y., the Public Health Service, Bethesda, Md., and Chas. Pfizer & Co., Inc., Brooklyn, N. Y.

TABLE 2

BLOOD SUGAR AND SERUM INORGANIC PHOSPHORUS CHANGES DURING GLUCOSE TOLERANCE TESTS IN ADULTS WITH AND WITHOUT CORTISONE ADMINISTRATION

Sex	Cortisone	Blood sugar, mg %						Serum inorganic phosphorus, mg %					
		30 min			2 hours			30 min			2 hours		
		Mean	±S D	No tested	Mean	±S D	No tested	Mean	±S D	No tested	Mean*	±S D	No tested
Healthy adults (21 to 60 years)													
M	0	50.3	21.0	31	7.0	18.9	63	−0.233	0.353	30	−0.463	0.705	63
M	200	48.8	24.4	39	12.8	21.5	59	−0.302	0.166	34	−1.024	0.467	57
F	0	35.8	27.0	34	9.0	18.1	69	−0.279	0.326	33	−0.572	0.440	68
F	200	37.9	25.7	48	12.2	22.0	65	−0.378	0.269	45	−1.072	0.449	65
Relatives of juvenile diabetics (21 to 60 years)													
M	0	49.9	23.4	51	21.4	35.1	60	−0.324	0.328	48	−0.567	0.463	56
M	200	44.9	16.1	16	8.6	24.1	24	−0.334	0.338	16	−1.020	0.469	25
F	0	43.2	23.8	53	11.4	24.9	67	−0.303	0.385	53	−0.502	0.494	64
F	200	56.4	13.5	19	17.[illegible]	18.7	28	−0.083	0.430	19	−0.776	0.511	29

* Difference between the bracketed pairs of mean values are statistically significant

Materials and Methods

Adult relatives of juvenile diabetics free of clinical symptoms of diabetes mellitus, apparently healthy adult male and female university personnel, healthy male dental students, and adult diabetics who did not require insulin received 1 75 gm of glucose per kilogram of body weight with or without the administration, 45 min earlier, of 200 mg of cortisone by mouth Blood

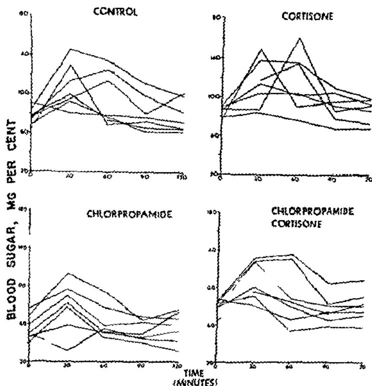


FIGURE 1 Blood sugar changes in healthy dental students during oral glucose tolerance test Glucose was administered at 1 75 gm kg of body weight with and without cortisone or chlorpropamide alone or in combination

for sugar and serum inorganic phosphorus analysis was withdrawn just prior to the administration of the glucose and at intervals thereafter, and was analyzed by methods in regular use in this laboratory [9] The studies with cortisone and chlorpropamide, alone or in combination, were limited to the dental students and to the adult diabetics

Results and Comments

Fasting blood sugar and glucose tolerance tests with and without cortisone in healthy adults with and without diabetic offspring TABLE 1 is made up of the mean fasting blood sugar and serum inorganic phosphorus values in healthy

university personnel and in relatives of juvenile diabetics in the 21 to 60 year age range with and without the prior administration of 200 mg of corti one 45 min earlier In addition to lower fasting blood sugar values in *the healthy university personnel* both male and female (indicated by the single asterisk) it is noteworthy that the administration of cortisone invariably produced lower levels of serum inorganic phosphorus (measured 45 min

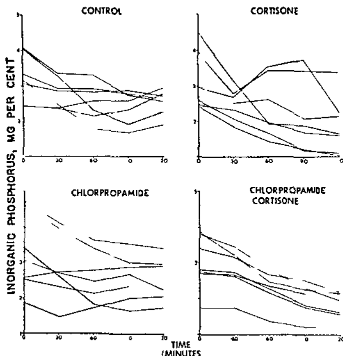


FIGURE 2 Serum inorgan c phosphorus changes n healthy dental students dur ng oral glucose tolerance test Gluc se was a lm n stered at 1 75 gm kg of bo ly we ght w th and without cort sone or chlorpropam de alone or in coml nat on

adults is in all probability an enhancement rather than a blockade of glucose utilization

The magnitude of the decline and the results of urinary analyses in selected patients indicated that the bulk of the observed decrease in serum inorganic phosphorus cannot be attributed to losses in urine

TABLE 2 shows the levels of blood sugar and serum inorganic phosphorus found one half hour and 2 hours following the administration *per os* of 1 75 gm of glucose per kilogram of body weight at zero time with and without 200 mg of cortisone given 45 min earlier In no instance did the steroid

TABLE 3

BLOOD SUGAR AND SERUM INORGANIC PHOSPHORUS CHANGES* DURING INTRAVENOUS GLUCOSE TOLERANCE TEST FOLLOWING ADMINISTRATION OF CORTISONE CHLORPROPAMIDE OR BOTH

	Cortisone	Chlorpropamide	Cortisone and chlorpropamide
Blood sugar	No change	Lower initial and final sugars with usual rise Blood sugar less than 60 mg per cent reached in 4 of 7 patients (Decreased gluconeogenesis or increased hepatic glycogenation without increased extrahepatic utilization)	No change
Serum inorganic phosphorus decrement	Greater (Increased gluconeogenesis enhanced insulin or both)	No change	Greater (In nondiabetic controls chlorpropamide did not alter cortisone effects)

* Compared to controls

affect the levels of blood sugar nor did it alter the serum inorganic phosphorus levels present at the 30 min point However, in each instance these amounts of cortisone greatly enhanced the usual hypophosphatemia usually doubling the magnitude of the decrease

it is evident
tal students
the hyper

glycemic response obtained with 1 75 gm of glucose per kilogram of body weight given at zero time On the other hand the prior administration of

moreover, that hypoglycemia, that is, values below 60 mg per cent developed in 4 of 7 subjects. Finally, the combination of cortisone and chlorpropamide in the dosages and at the time cited (FIGURE 1) canceled the general lowering of the blood sugar curves noted with chlorpropamide alone restoring them

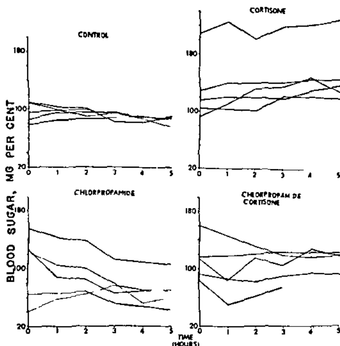


FIGURE 3. Blood sugar changes during oral glucose tolerance test. Glucose administered at 1.75 gm/kg of body weight in diabetics with and without cortisone or chlorpropamide alone or in combination.

to the levels seen in the control studies or in those made with cortisone alone certainly the absolute hypoglycemia seen with chlorpropamide alone did not develop.

From FIGURE 2 it is evident that cortisone administration at minus 45 min increased the hypophosphatemia that is a regular feature of such intravenous glucose tolerance tests in nondiabetic subjects. In the studies with chlorpropamide alone (FIGURE 2), in the majority of which the blood sugar reached absolute hypoglycemic levels the serum inorganic phosphorus did not decrease beyond the range seen in the control observations and certainly did not fall as low as in the cortisone experiments. In the combination of

cortisone and chlorpropamide (FIGURE 2) the serum inorganic phosphorus changes resembled those seen with cortisone The changes in the blood sugar and the serum inorganic phosphorus under these four circumstances are summarized in TABLE 3

The greater degree of hypophosphatemia in the cortisone studies suggests that this steroid increased the utilization of glucose by anaerobic glycolysis, by discharging insulin, prolonging or potentiating insulin action or by

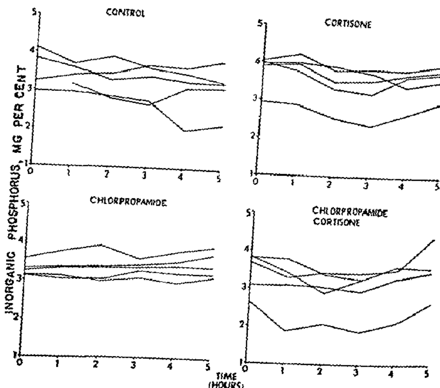


FIGURE 4 Serum inorganic phosphorus changes during oral glucose tolerance test Glucose was administered at 1 75 gm /kg of body weight in diabetics with and without cortisone or chlorpropamide alone or in combination

increasing cell permeability Increased gluconeogenesis could have occurred and potentiated this hypophosphatemic response by providing an increased glucose load

The lower blood sugar levels at zero time, throughout the course of the glucose tolerance tests and at their end, suggest that chlorpropamide decreased gluconeogenesis or enhanced the supplies or action of insulin or both However the fact that the serum inorganic phosphorus changes were no greater than those seen in the control studies argues for the former as the predominant or sole action

Cortisone and chlorpropamide effects in diabetics FIGURES 3 and 4 show the blood sugar and serum inorganic phosphorus levels in diabetics during several samplings for five hours with or without the prior administration of cortisone or chlorpropamide alone or in combination

In contrast to the shorter studies in the healthy university personnel in the relatives of diabetics and in the healthy dental students the blood sugar levels tended to rise when cortisone was given alone (FIGURE 3) However the same trend to hypophosphatemia noted in the three other groups was observed (FIGURE 4) Chlorpropamide alone tended to lower the blood sugar without decrease in the inorganic phosphorus (FIGURES 3 and 4)

In the chlorpropamide plus-cortisone studies it appears that chlorpropamide tended to produce an initial lowering of the blood sugar in three of the subjects (FIGURE 3) this was maintained in only one The serum inorganic phosphorus changes (FIGURE 4 assumed the pattern seen with cortisone alone

Under the conditions of these experiments chlorpropamide partially modified the hyperglycemia of cortisone administration probably by suppressing gluconeogenesis Since the serum inorganic phosphorus changes of the cortisone experiments were not accentuated it may be argued that chlorpropamide did not exert any significant action on any endogenous insulin that these patients may have had Alternatively it is possible that a slight enhancement was evoked just sufficient to offset the decrease resulting from the lesser degree of gluconeogenesis

Summary and Conclusions

Fasting blood sugar and glucose tolerance tests with and without cortisone in healthy adults and in relatives of juvenile diabetics The administration of 200 mg of cortisone 45 min before an oral glucose tolerance test (1 75 gm kg of body weight) lowered the fasting and the 2 hour levels of serum inorganic phosphorus without discernible change in the control blood sugar concentrations or increments This suggests that under the conditions of these experiments, in nondiabetic subjects this steroid enhanced the utilization of glucose by anaerobic glycolysis as reflected by significant increases in the usual hypophosphatemia This could represent increased gluconeogenesis increased secretion, intensification or prolongation of insulin action or increased cell permeability to glucose alone or in combination These findings argue strongly against a peripheral blockade of insulin action in these experiments

Action of chlorpropamide upon cortisone hypophosphatemia Chlorpropamide administered to healthy nondiabetic male dental students 45 min before an intravenous glucose tolerance test did not modify either the usual increment in blood sugar or the usual decrement in serum inorganic phosphorus although the absolute levels of blood sugar at the start during and at the end of the tolerance test were significantly lowered and hypoglycemia appeared in the majority

The failure to observe a further decrease in the serum inorganic phosphorus during these glucose tolerance tests suggests that the chief mode of action of the chlorpropamide was decreased gluconeogenesis or increased removal of extracellular glucose to form hepatic glycogen This is concluded since

both of these processes are known to lower the blood sugar without segregation of serum inorganic phosphorus

Our observations suggest that in the quantities and under the conditions employed cortisone cancels the hypoglycemic effect of chlorpropamide presumably by overriding the inhibition of gluconeogenesis by the latter but that chlorpropamide does not interfere with the cortisone enhanced hypophosphatemia

Blood sugar and serum inorganic phosphorus changes indicate that cortisone when first given to healthy subjects enhances rather than decreases the net disposal of glucose by processes that preempt serum inorganic phosphorus

Cortisone and chlorpropamide effects in diabetics In adult diabetics not requiring insulin 200 mg of cortisone given *per os* at minus 45 min tended to raise the venous blood sugar levels during several samplings for 5 hours while chlorpropamide had the opposite effect When the two were administered together the blood sugar levels dropped initially in 3 of 5 subjects, but the decrease was maintained in only 1 of these These findings suggest that, in the amounts given chlorpropamide could not completely overcome the hyperglycemic action of cortisone

Insofar as the serum inorganic phosphorus levels are concerned, no changes were observed with chlorpropamide while with cortisone or with cortisone and chlorpropamide a tendency to decreases was noted This again indicates that under these experimental conditions chlorpropamide did not cancel this action of cortisone

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INFLUENCE OF CHLORPROPAMIDE UPON POSTPANCREATECTOMY AND SPONTANEOUS DIABETES*

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Despite the current uncertainty regarding the mechanism of the hypoglycemic effect of the aryl sulfonylureas the observation of their failure to lower the blood sugar in severe alloxan diabetes in animals[1] and in depancreatized animals[2] and man[3, 4] remains unchallenged Although negative results were anticipated because of the structural similarity of chlorpropamide to carbutamide it was considered important to observe the action of the newer agent in pancreatectomized patients

Two patients in whom complete pancreatectomy had been performed by Porter and his associates[5] were selected One was a 60 year old woman 6 years after the operation the other a 55 year old man 16 months after removal of the pancreas In both surgery had been performed for pancreatic carcinoma Neither had evidence of recurrent or persistent tumor at the time of these studies

analyses tests of liver function (cephalin flocculation thymol turbidity serum bilirubin serum alkaline phosphatase serum cholesterol and serum protein electrophoresis) and measurements of phenolsulfonphthalein excretion and blood urea nitrogen Blood sugar levels were determined by the method of Folin and Wu at frequent intervals Those obtained 3 hours after breakfast are shown in FIGURES 1 to 3 Urinary glucose was estimated 5 times daily by the qualitative Benedict s method the mean daily value is shown in the figures Serum chlorpropamide (CPA) concentrations were determined by the ultraviolet spectrophotometric method developed by Toolan and Wagner[6]

Following the initial period chlorpropamide was administered in a dose of 1 gm daily and the insulin dose was simultaneously reduced from 20 to 10 U of NPH in the first patient M B (FIGURE 1) During the next 3 days glycosuria increased markedly and the patient developed thirst and polyuria The dose of chlorpropamide was raised to 2 gm on the fourth day after which it was discontinued owing to the appearance of nausea and vomiting The serum chlorpropamide level at this time was 30 mg 100 ml The first day

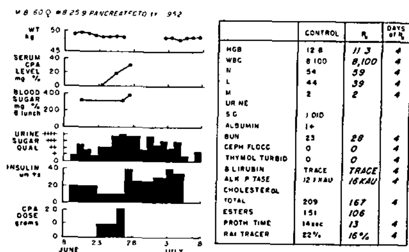
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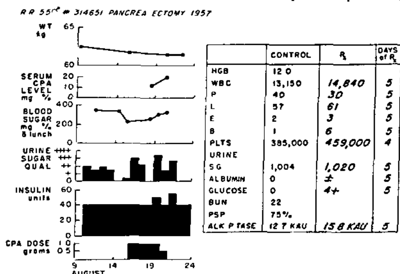
amide daily after the control period The previous dosage of 40 U of NPH

* The work reported in this paper was supported in part by a grant from Chas Pfizer & Co Inc Brooklyn N Y



	CONTROL	Rx	DAYS of Rx
HGB	12 8	11 3	4
WBC	8 100	8,100	4
N	54	59	4
L	44	39	4
M	2	2	4
UR NE			
S G	1 010		
ALBUMIN	1+		
BUN	23	28	4
CEPH FLOCC	0	0	4
THYMOL TURBID	0	0	4
B LIRUBIN	TRACE	TRACE	4
ALK P TASE	12 1 KAU	16 KAU	4
CHOLESTEROL			
TOTAL	209	167	4
ESTERS	151	106	
PROTH TIME	14 sec	13	4
RAI TRACER	22%	16%	4

FIGURE 1 Response to chlorpropamide of patient M B six years after pancreatectomy



	CONTROL	Rx	DAYS of Rx
HGB	12 0		
WBC	13,150	14,840	5
P	40	30	5
L	57	61	5
E	2	3	5
B	1	6	5
PLTS	385,000	459,000	4
URINE			
S G	1,004	1,020	5
ALBUMIN	0	±	5
GLUCOSE	0	4+	5
BUN	22		
PSP	75%		
ALK P TASE	12 7 KAU	15 8 KAU	5

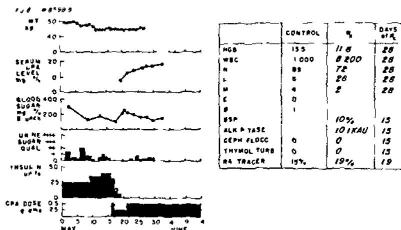
FIGURE 2 Response to chlorpropamide of patient R R sixteen months after pancreatectomy

insulin was continued because of persistently elevated blood sugar levels Despite the addition of chlorpropamide, with serum concentrations to 20 mg /100 ml, the blood sugar rose and glycosuria increased during the experimental period

Laboratory studies (FIGURES 1 and 2) indicated the presence of a mild neutropenia of questionable significance, which increased in R R after chlorpropamide administration Minimal albuminuria also was observed in this patient after treatment with this agent The fall in serum cholesterol

in M B is difficult to evaluate because of major alterations in food intake during part of the period The serum alkaline phosphatase concentrations increased in the 2 patients from 12 1 to 16 and from 12 7 to 15 8 King Armstrong units (normal, 12 or less), respectively

Chlorpropamide was also given to 6 patients with spontaneous, stable diabetes that began after 30 years of age Among them were 5 women and 1 man ranging in age from 58 to 81 years and, in insulin requirement, from 20 to 60 U Insulin was discontinued and 0 5 to 1 gm daily doses of chlorpropamide were given for periods of 2 weeks to 4 months In 4 members of



	CONTROL	℞	DAYS of ℞
HGB	13 5	11 8	28
WBC	1 000	8 200	28
N	89	72	28
L	5	26	28
M	4	2	28
E	0		
B	1		
BSP		10%	15
ALK P TASE		10 IKAU	15
CEPH FLOCC	0	0	15
THYMOL TURB	0	0	15
RA TRACER	15%	19%	19

FIGURE 3 Response to chlorpropamide of a patient with spontaneous diabetes

this group, blood sugar concentrations were maintained at 200 mg 100 ml or less 3 hours after breakfast FIGURE 3 summarizes the observations in a patient whose blood sugar levels were satisfactorily controlled by chlorpropamide alone This agent replaced 30 U of globin insulin in an obese 77 year-old woman following an unsuccessful trial of tolbutamide

Of the 2 patients

be substituted successfully for 40 U of protamine zinc insulin The other a 68 year old man well regulated on 20 U of protamine zinc insulin, had also failed to respond to tolbutamide Although an initial course of chlorpropamide restored the blood sugar values to essentially normal levels a second trial of this agent after 1 week on tolbutamide was ineffective in preventing hyperglycemia

that in neither was control of the blood sugar adequate prior to chlorprop

amide administration Parallel with previous observations on other aryl sulfonylureas Root has reported the potentiation of insulin action by chlorpropamide in 2 totally depancreatized dogs Such an effect was not found in the present study on 2 human subjects, although it should be noted that the agent was given for much shorter periods In one of Root s dogs the serum alkaline phosphatase increased and a fatty liver was found at autopsy The significance of the small increases in alkaline phosphatase levels in the 2 patients reported here requires further evaluation In the 6 patients with spontaneous diabetes no disturbance of liver or kidney function was seen during chlorpropamide treatment nor was there evidence of alteration of the bone marrow or of allergic reaction

Effect of Chlorpropamide upon Plasma Unesterified Fatty Acid Concentrations

The importance of plasma unesterified fatty acids (UFA or NEFA) in lipid transport and their relationship to carbohydrate metabolism have been

TABLE 1
PLASMA UNESTERIFIED FATTY ACID LEVELS FOLLOWING ORAL CHLORPROPAMIDE*

Time (min)	Serum chlorpropamide				Blood sugar				Unesterified fatty acids			
	Patient				Patient				Patient			
	a†	b	c	d	a	b	c	d	a	b	c	d
	(mg 100 ml)				(mg 100 ml)				(mEq /l)			
0	0	0	0	0	142	130	84	217	0 50	0 36	0 33	0 67
60				2 9	132	114	82	196	0 34	0 24	0 36	0 47
165	24				112	84	61	164	0 27	0 16	0 31	0 33
225		25	20		108	93	63	115	0 34	0 35	0 36	0 24
300		24	25	18		92	59	70		0 48	0 43	0 50

* Patients a b and c were given 3 gm of chlorpropamide patient d received 2 gm
† Patient a took NPH insulin 10 U daily until 48 hours prior to this experiment patients b c and d required no insulin

demonstrated by Dole [8] Gordon [9] and others Elevated levels in diabetic patients in the postabsorptive state and during ketosis have been demonstrated by Bierman [10] The high UFA concentrations found in acidosis fall sharply in response to insulin The hypoglycemic effect of tolbutamide is accompanied by a decline in plasma UFA concentrations [11]

In the present study 4 patients with mild diabetes were given a single 2 or 3 gm oral dose of chlorpropamide after a 9 or 14 hour period of fasting Only 1 member of the group required insulin, this was discontinued 48 hours prior to the study Plasma levels of sugar, chlorpropamide, and UFA were

letermined at interval for 5 hours The method of Cor lon as u e l fo UFA estimat on

The result are l ted n T BLE 1 The blood ugar lecrease l n all patient a the serum hlorpropam le level ro e to 18 mg 100 ml or more The UFA con entration fell n 3 pa ent as un hanged n 1 and at the end o the study ro e n all

C l sio

Chlorpropam de l ke arbu am le and tolbutam de an effe t ve ora hypo lycem agent n ome patient with l abetes Fa lure of chlorpr p am de to p o lu e a de rea e n the blood ugar le el n the ab ence of the pancrea a ob er ed n t o pat en A mall ncrea e n erum alkal ne pho phata e level wa f un l n both pancreatectom zed pat ents after treat ment h hlorp opamide for b ef per od Pla ma une ter f ed fatty ac leve de rea ed after chlorpropam le as they do after n ul n a lm n strat on

Ack o ledgme t

We are ndebted for echn cal ass tan e to Barbara Jone and Em ly B l ell and al o t Margare Ha thorne of the Metabol m Ward of the Pre by ter an Ho p al

Adde d

A e p e a a on o h s pa e 2 a ien w h pon an ous iabe e o xd g n ican e a ons of se um a ka ne pho ha ase on en a ns wh e e ng 0 gm doses of hl p o am de da In one w h d a a o hlorp opam de was fo on p m e u n he alka ne pho ha ase o no ma e els The o he pa ien af e e n ng hlo propam de n 0 25 gm dose de lo e a se e esk n eru n an la holang o he a w h eco e n l mon h

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Discussion of the Paper

HENRY RICKETTS (*Department of Medicine, University of Chicago School of Medicine, Chicago, Ill.*): I think it is still quite mysterious that the human being without a pancreas and maintained on insulin fails to show a depression of blood sugar when sulfonylureas are added to the regimen, in contrast to the totally depancreatized dog in which (as shown by a number of workers including ourselves) the addition of sulfonylureas to suboptimal doses of insulin results in a clear-cut depression of blood and urinary glucose. I have yet to hear why the human being responds differently from the dog in this respect, and I wish we could have the answer.

When speaking of the effects of sulfonylureas in depancreatized people or animals, we should be careful about our use of the term "potentiation of insulin." Insulin potentiation implies accentuation of the effects of insulin; this is not quite true in depancreatized dogs because, as we have demonstrated in a work thus far unpublished, in the totally depancreatized dog maintained on suboptimal amounts of insulin and receiving chronically added sulfonylureas, there is a clear reduction in blood and urinary sugar with *no* change of nitrogen excretion. Since the addition of 2 U. of insulin to the regimen in these animals produces the expected decline in blood and urinary sugar and, now, a clear diminution of nitrogen excretion, I do not think we can refer to the sulfonylurea effect as insulin potentiation. I suspect that it is not and I suspect also, if our evidence is valid, that one must conclude that in the dog without a pancreas the sulfonylurea effect occurs in other tissues for whose integrity a small minimum of insulin is necessary.

THE EFFECTS OF CHLORPROPAMIDE ON ENDOCRINE FUNCTION IN PATIENTS WITH DIABETES MELLITUS AND ITS EFFECTS IN OTHER ENDOCRINE DISORDERS*

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The continued widespread use of tolbutamide for the treatment of diabetes mellitus has left little doubt about either the efficacy of action or the acceptance by both the physician and patient of this oral agent for the treatment of this disease Similar drugs are destined to undergo investigation, and certain of these will find a place as competitors with, or substitutes for, tolbutamide

In order to compete with tolbutamide a compound must be at least as potent a hypoglycemic agent and equally innocuous from the standpoint of toxicity Additional attributes of such a drug might include equal effectiveness at lower dose ranges or blood levels less expense to the patient or usefulness as alternate therapy in patients known to be hypersensitive or unresponsive to tolbutamide

It has been the aim of our study to accumulate data on chlorpropamide regarding each of these points Preliminary results indicate that chlorpropamide is effective as an oral hypoglycemic agent in the same type of diabetic patient that responds to tolbutamide and that dosages of approximately one third the magnitude of the latter drug result in equal or higher serum levels Additional data suggest that hypoglycemic reactions are more common with chlorpropamide, but in short term usage few manifestations of toxicity to vital organs were observed As with other sulfonylurea derivatives slight inhibition of thyroid function may occur but this does not appear to be clinically significant

Methods and Results

Fifty four patients were selected for a trial of chlorpropamide therapy because as a group they characterized the diabetic most likely to respond to a sulfonylurea hypoglycemic agent as regards age, sex, body type, duration of diabetes and daily insulin need The group included 13 recently discovered diabetics who could not be controlled with diet, 13 individuals previously controlled with insulin and 28 patients who had been treated with tolbutamide (TABLES 1 2 and 3)

Chlorpropamide was administered orally in doses ranging from 0 2 to 1 0 gm daily, and frequently a priming dose of 1 or 2 gm was given Blood sugar levels were obtained daily in both the fasting and postprandial states

* Work reported in this article was supported in part by Research Grant 2A 5118 (C) and (CS) from The National Institute of Arthritis and Metabolic Diseases Public Health Service Bethesda Md by the Comly Coleman Fund of the Ohio State University, Columbus Ohio, and by Chas Pfizer & Co , Brooklyn, N Y

Table 1
Method of Study of Hypoglycemic Effect of Chlorpropamide

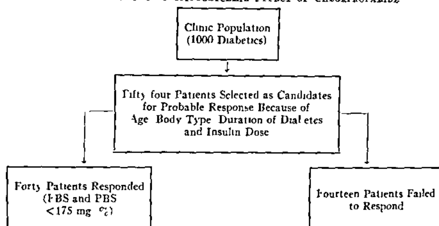


Table 2
Characteristics of Responsive and Nonresponsive Patients

Factors	Responsive (40 patients)	Nonresponsive (14 patients)
Mean age (years)	60	56
Percentage female	80	71
Percentage more than 20 lb overweight	75	64
Duration of diabetes (years)	5.6	6.4
Percentage with history of ketosis	3	29
Mean daily insulin (units)	11	25

Table 3
Treatment Before Chlorpropamide

	Responsive (40 patients)	Nonresponsive (14 patients)
Controlled on tolbutamide	13	0
Uncontrolled on tolbutamide	10	5
Uncontrolled on diet alone	10	3
Controlled with insulin	7	6

in hospitalized patients and weekly or biweekly in outpatients Patients were counted as responsive if all blood sugar levels fell below 175 mg per cent and remained below this level Chlorpropamide treatment was continued for at least 7 days in nonrespon ive patients and usually until the serum level of the drug approached or exceeded 20 mg per cent Treatment was continued in all patients who responded except in instances where untoward reactions developed

Forty of the 54 patients met the criteria for response and 14 failed to do so As a group those responding were slightly older more obese and had a

TABLE 4
RESULTS OF CHLORPR PAMIDE TREATMENT IN PATIENTS FORMERLY TREATED WITH TOLBUTAMIDE

Patients	Tolbutamide period		Chlorpropam de period		
	Mean FBS (mg)	Daily se (gm)	Mean FBS (mg)	Daly dose (gm)	1 607 level (mg)
Group 1					
Response to both agents 13 patients	135 (110-164)	1 54 (1 2	110 (60 162)	0 45 0 2 1 0	13 (20
Group 2					
Response to chlorpropamide *only* 10 patients	250 (200–380)	1 60 1 3	123 80-160)	0 57 (0 2 1 0	18 17 20)
Group 3					
Unrespons ve 5 patients	243 (215 2 0)	1 50 (1 2)	229 215 2 0)	0 45 (0 2 0 5)	22 (16 29

shorter duration of the diabetic state than those who did not respond These differences between the groups were slight and statistically insignificant however the mean pretreatment daily insulin dose was 11 U in the respon sive group and 25 U in the nonresponsive group and only 1 of the 40 patients in the former group gave a history of past ketoacido is while 4 of 14 in the latter group had such a history

Twenty eight patients who had taken tolbutamide were subjected to a trial of chlorpropam de in order that blood sugar responses and magnitudes of dosage of the 2 drugs m ght be compared (TABLE 4) Chlorpropamide in daily do es of 0 2 to 1 0 gm was substituted for tolbutamide in 13 patients (Group 1) who previously had re ponded in an optimal fashion to the latter drug The degree of control with chlorpropamide in a mean dose of 0 45 gm daily compared favorably to that obtained with the daily administration of 1 54 gm of tolbutamide, and no patient who previously had been controlled

with tolbutamide was found to be resistant to chlorpropamide Serum levels of chlorpropamide averaged 13 mg per cent in this group

Fifteen additional patients had been taking 1 0 to 3 0 gm of tolbutamide daily, but displayed fasting blood sugar levels of 200 to 380 mg per cent and could be classified as relatively resistant to tolbutamide at this dosage level When chlorpropamide was substituted, 10 of these individuals (Group 2 showed a fall in mean fasting blood sugar levels from 250 to 123 mg per cent and each met our criteria for response At the time of the favorable response the dose of chlorpropamide ranged from 0 2 to 1 0 gm daily, and serum levels of the drug averaged 18 mg per cent However, in 5 subjects (Group 3) the fasting blood sugar levels fell only slightly, from a mean of 243 to 229 mg

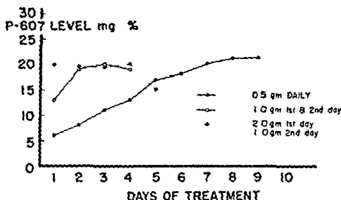


FIGURE 1 Effect of initial chlorpropamide dosage on chlorpropamide blood levels *Solid curve with closed circles represents data from 4 patients treated with dosages shown* the solid curve with open circles 2 patients and the broken curve 2 patients

per cent as chlorpropamide in daily doses of 0 25 to 0 50 gm was substituted for an average dose of 1 5 gm of tolbutamide Serum levels of chlorpropamide ranged from 16 to 29 mg per cent in these unresponsive patients

It became apparent early in the study that some correlation existed between magnitude of dosage serum levels of the drug and response Accordingly, initial daily doses ranging from 0 5 to 2 0 gm were administered to small groups of subjects and the relationship between the serum levels obtained and the lapse of time needed for attainment of satisfactory blood sugar levels was noted (FIGURE 1) When a dosage schedule of 0 5 gm daily was employed, about 4 days were necessary to achieve those serum concentrations of the drug that were found to be optimally effective This dose resulted in progressive elevation of serum levels for 9 to 10 days, after which constant levels were maintained When larger initial doses were used, higher serum levels were achieved much more rapidly With either method optimal control of hyperglycemia was attained only after 4 to 12 days (average 6 days)

Blood levels of chlorpropamide obtained at random in a group of 20 patients maintained for 1 to 20 weeks on a daily dose of 0 5 gm were found to range from 16 to 20 mg per cent (FIGURE 2) In patients in whom the drug was

discontinued, serum levels as high as 5 mg per cent persisted as long as 72 hours

Endocrine function The effect of chlorpropamide upon thyroid function was assayed by determination of serum protein bound iodine (PBI) levels,

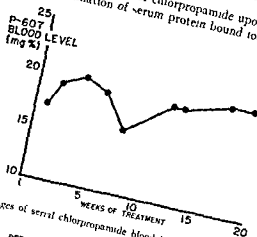


FIGURE 2 Averages of serial chlorpropamide blood levels of 20 patients maintained on 0 5 gm daily

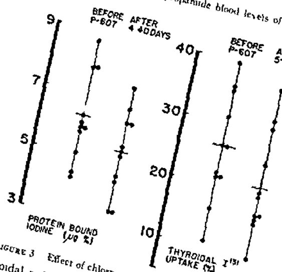


FIGURE 3 Effect of chlorpropamide on thyroid function tests

24 hour thyroidal radioiodine uptakes, and serum cholesterol values, both during the pretreatment period and after 4 to 50 days of therapy and, subsequently, at random intervals Comparison of these values suggests some inhibition of thyroid function, as the mean PBI fell from 6 2 to a level of 5 4 μg per cent, and the mean uptake fell from 27 to a value of 22 per cent (FIGURE 3) This effect of a sulfonylurea derivative has been described by

an improvement in diabetic control rather than an alteration in thyroid function although a direct effect on lipid metabolism cannot be excluded. In 2 patients with concomitant thyrotoxicosis and hyperglycemia, chlorpropamide proved to be an effective hypoglycemic agent.

The relationship of chlorpropamide therapy to adrenal cortical function was studied by quantitative measurement of daily 24 hour urinary excretion

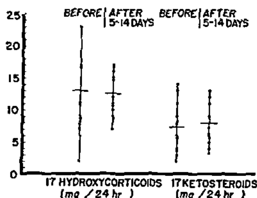


FIGURE 4. Effect of chlorpropamide on urinary steroid excretion

of 17 hydroxycorticoids and 17 ketosteroids. Values for each patient were averaged individually and control levels were compared with those obtained during treatment. No changes attributable to therapy were noted. Corticoids averaged 13.5 mg per day before therapy and 13.0 mg per day during therapy; ketosteroids averaged 8.0 mg per day in the control period and 8.5 mg per day during treatment (FIGURE 4). Each of 3 subjects with Cushing's syndrome due respectively to adrenal cortical hyperplasia, adrenal cortical adenoma, and exogenous administration of 6 methyl prednisolone displayed significant lowering of blood sugar and became normoglycemic with chlorpropamide treatment. This response differs from that reported by others who failed to observe such an effect when using tolbutamide.*

A 25 year old juvenile diabetic who was also treated had undergone a surgical hypophysectomy in the hope that his severe progressive retinitis proliferans might be controlled. Hypophysectomy resulted in a decrease of his daily insulin requirement from 80 to 14 U of NPH. The daily admin

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ion

Reactions to chlorpropamide. Numerous tests were ... the influence of chlorpropamide on the functions of the liver, kidney, and bone marrow (TABLE 5). Such determinations were made prior to the inception of chlorpropamide therapy and were repeated after 5 to 10 days and later

at random intervals throughout the period of study Averages and ranges of values obtained before therapy were compared to the mean determinations made during treatment

No significant alterations in liver function were evident from the comparison of values of thymol turbidity serum alkaline phosphatase serum

TABLE 5
TESTS OF LIVER KIDNEY AND BONE MARROW FUNCTION

Test	No of patients	Values before chlorpropamide	Values after chlorpropamide
Thymol turbidity (units)	16	5 (0 20	9 4 (0–40)
Serum alkaline phosphatase (SJR units)	13	6 8 (4 14	6 3 4 16)
Serum bilirubin (direct indirect mg)	14	0 3 0 3 0 1 0 1–0 (0 2)	0 2 0 1 (0 1 0 1–0 5 0 4)
Prothrombin concentration (%)	9	83 52 100)	81 42 100
Albumin globulin (gm %)	9	4 3 3 3 2 6 1 –4 8 4 4	4 1 3 5 2 5 2 6–4 6 4 0
Bromsulphalein retention (percentage after 45 min)	10	3 5 (0 14	2 5 0–8)
Cephalin flocculation (units)	10	trace 0 2+	trace 0 1+)
Serum glutamic oxaloacetic transaminase (units)	10	19 7 (11 2)	29 8 7 103)
Serum creatinine (mg %)	9	1 4 1 0 1 7	1 4 1 0–1 9)
Blood urea nitrogen (mg %)	34	16 8–31	18 (8 32
Serum uric acid (mg %)	9	3 6 3 0–4 4	3 3 0 4 7
White blood cell count (per cu mm)	25	7400 4 000 20 000	6400 3 500–10 400
Neutrophils (percentage)	24	61 (43 7	60 35 74
Hemoglobin (gm %)	21	13 9 12 16)	13 4 11 16

bilirubin prothrombin concentration serum albumin and globulin Bromsul [illegible]

4 categories Of the 54 patients to whom the drug was given 9 (17 per cent) manifested undesirable effects Three individuals all of whom were lean mild diabetics experienced classic hypoglycemic responses These episodes

occurred in both the late postprandial and fasting states and were rapidly controlled by glucose administration. Decrease in the maintenance dose abolished these reactions. Two subjects complained of burning midepigastric distress following ingestion of the tablets. Each had registered similar complaints before being placed on therapy but felt that the drug exaggerated the discomfort. This complaint was easily controlled by administering antacids with the drug or by giving the drug with meals. Many patients with varying degrees of severe cerebral arteriosclerosis were treated with chlorpropamide. In 2 of these cerebral vascular accidents were observed during treatment. In another episodes of accentuated mental confusion occurred. In none was actual hypoglycemia documented although this would appear to be the most likely explanation for their symptoms.

For the most part no alteration of hepatic function could be attributed to the drug. However, in one patient with biopsy proved hepatic cirrhosis slight enlargement of the liver and increase in thymol turbidity titers occurred during chlorpropamide therapy. Similar episodes had occurred during periods when the drug was not being given. There were no manifestations of dermal hypersensitivity to the drug.

Summary and Conclusions

Chlorpropamide was given to 54 selected diabetic patients and an adequate response was obtained in 40 of these. As a group the patients tended to be mild diabetics requiring little or no insulin, were obese and middle aged or older.

Ten of 15 patients unresponsive to tolbutamide responded optimally to chlorpropamide. No patient responsive to tolbutamide failed to respond to chlorpropamide.

The effective maintenance dose ranged from 0.2 to 1.0 gm daily and resulted in serum levels of 13 to 20 mg per cent. A priming dose raised the serum levels more rapidly but failed to hasten the hypoglycemic response.

Slight depression of thyroid function as determined by the usual tests was observed but no clinical manifestations of thyroid deficiency or enlargement of the thyroid gland occurred.

No alterations of adrenal cortical, renal, hematopoietic or cardiac function were observed. Liver function remained normal except in 1 patient with known hepatic cirrhosis.

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Two patients with thyrotoxicosis and 3 with Cushing's syndrome associated with hyperglycemia became normoglycemic with chlorpropamide treatment.

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Discussion of the Paper

Carlos P Lamar (*Miami Fla*) I propose to make a short comment on another aspect of the side effects of sulfonylureas that I do not believe has been mentioned in these pages

Hamwi and others reported on the number of patients who showing late developing lack of respon e or resistance to sulfonylureas were necessarily returned to insulin therapy We have 7 patients who were originally selected as ideally suited for this type of therapy and who apparently responded well for periods of 2 to 7 months after which it was necessary to return them to insulin The original daily doses of insulin required for maintenance of adequate diabetic control for periods of several years had ranged between 20 and 30 U Following the use of tolbutamide and carbutamide in this group, the amount of insulin required to obtain adequate diabetic control ranged from 60 to 90 U , or 3 times the pretolbutamide or carbutamide dose level These individuals have been observed now for periods of 6 months to almost 2 years and although their insulin requirements are declining gradually, they still require 100 per cent more insulin than they did before

With respect to Hamwi s cases of cerebral arteriosclerosis with manifestations of clinical hypoglycemia in which he was unable to substantiate the presence of chemical hypoglycemia I suggest a very simple therapeutic test that we have used very successfully for quite some time administer sugar intravenously even if the individual has a blood sugar level of 250 to 300 mg per cent or more These figures do not mean that that individual who probably has a decreased transfer or osmosis through the cerebral cell membrane is not hypoglycemic the proof of hypoglycemia lies in the fact that the administration of glucose under these circumstances is followed by a dramatic correction of the clinical hypoglycemic symptoms An adjustment in the treatment of these individuals to a less rigid adherence to the concept of so called normal glycemic levels would help them and perhaps might be a factor in the prevention of those cerebral vascular accidents mentioned as of obscure origin in this and other individuals

CLINICAL EXPERIENCE IN THE USE OF CHLORPROPAMIDE IN THE TREATMENT OF DIABETES MELLITUS*

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Metabolic Endocrine Division National Institute of Nutrition Mexico D F Mexico

It is our purpose in this paper to describe our clinical experience with chlorpropamide in the management of the adult and juvenile diabetic patient

In fifty unselected subjects from the Diabetic Clinic of the Nutritional Disease Hospital in Mexico D F, Mexico, the influence of chlorpropamide has been studied with particular reference to changes in hyperglycemia and the general health of the patient In a few cases, studies of thyroid function were carried out

Controls

The hypoglycemic effect of the compound was first tested in 10 healthy normal young adults who were used as a control group These patients were given glucose tolerance tests using a standard 100 gm oral dose, and measuring blood sugar every hour for 5 hours This study was repeated on the same subjects 3 to 6 days later, each patient receiving a test dose of chlorpropamide (1 gm) 1 hour before the glucose ingestion

Results The results of this study are shown in FIGURE 1 There is a definite flattening of the curve after chlorpropamide It is interesting to note the significant depression in the fasting blood sugar values during the test in 6 of the 10 subjects

Diabetic Patients

Material and methods In this group of 50 cases, diabetes was of the adult or stable type in 45 and juvenile or brittle" in 5 The duration of the disease ranged from 3 months to 19 years, with an average of 6½ years

Most patients were between 40 and 50 years old, the ages ranging from 16 to 70 years The age distribution is shown in TABLE 1 Forty four cases had vascular manifestations of diabetes, among the 50 cases there were 30 other complications as shown in TABLE 2 Before the beginning of the study, 8 patients were on diet alone 33 were receiving insulin 5 were on carbutamide or tolbutamide and 4 were receiving no treatment (TABLE 3)

All patients were kept on a constant calculated diet during a control period and throughout the study The diet components were measured in terms of familiar household serving practices (ambulatory cases), or were supplied by the dietetic kitchen (hospitalized cases)

Chlorpropamide therapy was usually started in divided doses of 750 mg to 1 gm in 24 hours The duration of treatment was 8 to 69 days

Patients who had failed to respond to diet alone or who were on other oral hypoglycemic agents received chlorpropamide as the only medication The dose of the drug was modified subsequently in each case, according to

* The work reported in this paper was aided by a grant from Pfizer de Mexico Mexico D F Mexico who supplied the chlorpropamide used

the initial response Chlorpropamide was given alone when daily insulin requirements were 20 U or less

A reduction of 75 per cent of the insulin dose was made in patients receiving more than 20 U per day, and chlorpropamide was given in the same fashion

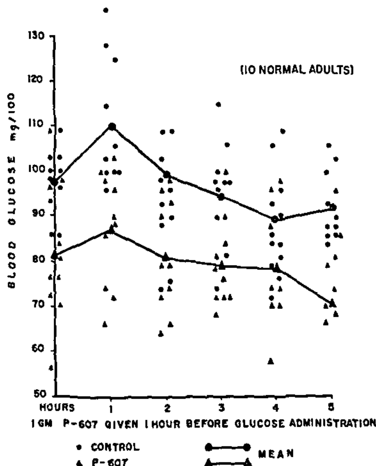


FIGURE 1 Changes in glucose tolerance test with chlorpropamide

described for the whole group Further reductions in insulin were made according to the response

Diabetic control was evaluated on the basis of glycosuria and glycemia Urine sugar was checked three times a day by the qualitative Benedict's method, and the results were recorded in the usual way from 0 to ++++ Blood glucose determinations were made by the Folin Wu method in the fasting state and, occasionally, after meals in the ambulatory cases

TABLE 1
AGE DISTRIBUTION OF PATIENTS

Age (years)	No of cases
10–19	2
20–29	3
30–39	2
40–49	15
50–59	21
60–69	5
70–79	2

TABLE 2
VASCULAR MANIFESTATIONS AND COMPLICATING DISEASES

Disorder	No of cases
Retinopathy	27
Nephropathy	5
Cataract	7
Neuropathy	18
Urinary infection	3
Gangrene	1
Arteriosclerotic heart disease	11
Tuberculosis	2 (1 inactive)
None	14

TABLE 3
DATA REGARDING PREVIOUS TREATMENT

Treatment	No of cases
Diet alone	8
Insulin	33
Tolbutamide	4
Carbutamide	1
None	4

Outpatient cases were followed twice weekly and close clinical observation was maintained Particular emphasis was placed on the detection of toxic manifestations In twenty seven cases, liver function tests were done prior to the treatment and were repeated about three weeks later These included the Bromsulphalein test and measurements of serum bilirubin, cephalin cholesterol flocculation, and thymol turbidity Complete blood counts were done before chlorpropamide in forty cases and repeated toward the end

of the therapeutic trial Because of the possibility of crystal[illegible] urinalysis was done in all cases

Results

According to the clinical and laboratory data [illegible] chlorprop-amide treatment the diabetic patients were placed [illegible] groups (Table 4)

TABLE 4
CRITERIA FOR CLASSIFICATION OF RES[illegible]

Classification	Characteri[illegible]
Good	At least three of the following a) Fasting blood sugar less than 130 mg b) Postprandial blood sugar less tha[illegible] c) Zero to 1 plus glycosuria d) Complete or almost complete [illegible] n require-ment
Fair	a) Fasting blood sugar less than [illegible] b) Postprandial blood sugar less th[illegible] c) One plus to 2 plus glycosuria d) At least 50 per cent reduction in insu[illegible] ment
Poor	All others

Good response group Constituting 56 per cent of the total patients in this group display at least 3 of the following characteristic (1) a fasting blood sugar of less than 130 mg per cent (2) a postprandial blood sugar less than 170 mg per cent, (3) zero to 1 plus glycosuria and (4) complete or almost complete substitution (75 per cent reduction) in insulin requirements

Fair response group Patients (22 per cent of total) in this group show (1) a fasting blood sugar of less than 150 mg per cent (2) postprandial blood sugar of less than 200 mg per cent (3) one plus to 2 plus glycosuria and (4) at least 50 per cent reduction in insulin requirements

Poor response group This group is constituted of all patients (22 per cent) not classified in the other groups

Using the above criteria, which are related entirely to the control of the hyperglycemic and glycosuric aspects of diabetes a good response was observed in 28 of 50 cases (56 per cent), a partial effect in 11 (22 per cent) and none in 11 (22 per cent)

Discussion

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an almost complete substitution (75 per cent) in 1

Two patients were on tolbutamide and 1 on carbutamide There was a

transitory period of hyperglycemia and intense glycosuria immediately following the substitution of tolbutamide for chlorpropamide in 1 case, but glucose levels promptly returned to normal within the following 48 hours

The nine remaining patients were without insulin or other hypoglycemic agents at the start of the study. They failed to respond to diet alone, but responded to chlorpropamide

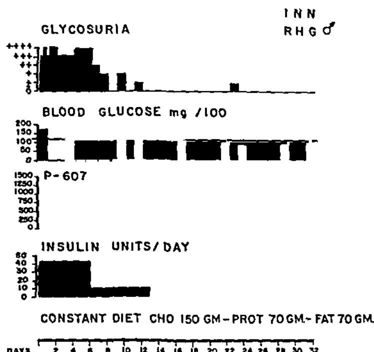


FIGURE 2

Fasting blood sugar values in the pretreatment period in the whole group ranged from 90 to 350 mg/100 cc, the average being 203 mg. After chlorpropamide administration, values varied from 79 to 161, the average being 110 mg/100 cc

The favorable response in the cases considered "good" occurred between the second and eighteenth days, the average at 5.5 days. In most patients, after controlling their diabetes, the average daily maintenance dose of chlorpropamide was 500 mg

An illustrative case is presented in FIGURE 2. The drug was well tolerated, but we noted slight hypoglycemia in three cases

Case 44 seemed to be a relative failure at the beginning, as there was a period of 17 days before glucose levels were controlled with 1 gm. of chlorpropamide and decreasing doses of insulin. Ten days later we were able to discontinue insulin and to control the patient with chlorpropamide (FIGURE 3)

In the fair response group, 7 patients were receiving insulin in doses that ranged from 20 to 80 U per day, the average being 43 U —that is, 15 U more than the mean of the good response group At the beginning of the study 1 patient was on tolbutamide, 2 were on diet alone (but their diabetes was uncontrolled) and 1 was receiving no treatment Chlorpropamide administration was maintained for an average of 22 days, with a range of 8 to 38 days The mean daily maintenance dose in this group was 770 mg

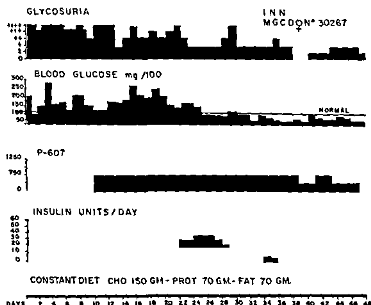


FIGURE 3

Average values for the fasting blood sugar dropped from 207 mg /100 cc to 152 mg /100 cc after chlorpropamide administration A typical case is presented in FIGURE 4

At the beginning of the study, 10 patients in the poor response group were receiving 25 to 60 U of insulin per day, the average being 37 U , and 1 was on tolbutamide but had not responded to this compound Our five patients [illegible]

propamide was given in a dose of 1 gm per day, the average duration of treatment was 12 6 days A case is presented in FIGURE 5

Also studied was the possibility of a correlation between the response to chlorpropamide and several factors such as the type of diabetes the presence

of obesity, the previous insulin requirements, and the duration of the disease The results are shown in FIGURE 6

The only positive correlation found was that between response and disease type, as no single case of juvenile diabetes responded to chlorpropamide treatment

Side effects and toxic manifestations Eleven patients complained of somnolence, weakness, and a "dizzy feeling" that were thought to be due to

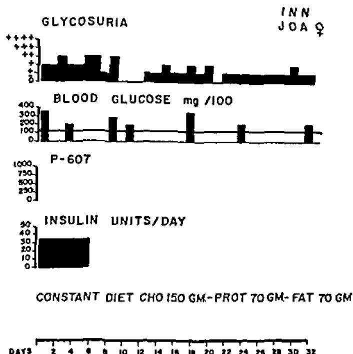


FIGURE 4

hypoglycemia, but glucose determinations failed to confirm this impression Dermatological lesions were seen in 3, of these, 1 was an exfoliative dermatitis that promptly subsided after discontinuance of therapy The other 2 patients had urticaria this disappeared in 1 in spite of continuing treatment and, in the remaining case after reducing the dose from 1 gm to 750 mg per day Four patients complained of constipation

No change in the liver function tests was shown in the 27 patients in whom such studies were done before and during chlorpropamide therapy

Aside from transitory reduction in granulocytes in 3 cases, no toxic effects on the hematopoietic system were observed

No crystalluria was found during the period of observation

Thyroid function, as demonstrated by uptake of I^{131} by the thyroid gland, was measured in 7 cases before the onset of chlorpropamide treatment, this was repeated 2 to 3 weeks later, no significant depression was observed

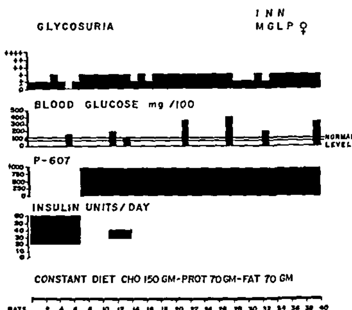


FIGURE 5

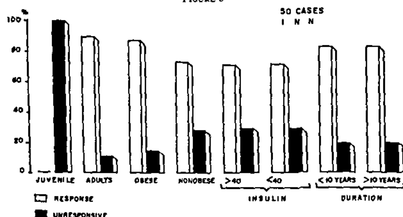


FIGURE 6 Response of diabetic patients to chlorpropamide

Summary and Conclusions

Chlorpropamide was administered to ten normal healthy adults and fifty diabetic patients in order to study its hypoglycemic activity a flattening of the glucose tolerance curve was seen after its administration

Fifty six per cent of the diabetic patients responded favorably to the

was obtained

No alterations in liver function or in the hematopoietic system were observed

Side effects included somnolence weakness and dizziness in eleven cases

Response does not seem to be related to the duration of the diabetes to the insulin requirements (within certain limits) or to deviations from normal weight

SUMMARY OF THE MONOGRAPH

Charles H. Best

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We all appreciate the fact that new evidence derived from the use of oral hypoglycemic agents has focused a searching light on the formation, liberation and action of insulin on a variety of metabolic processes and on diabetes and its treatment. We acknowledge again our debt to Loubatières[1] for his pioneering work and our indebtedness to our German colleagues[2-4] for their great and essential chemical and clinical contributions to this rapidly expanding field. I was just beginning to take a scientific interest in metabolism when the work on the hypoglycemic effect of guanidine was published in 1918.[5] We lived through the studies and clinical use of Synthalin,[6] Myrtillin and many other disappointing hypoglycemic agents.[6] However, there is no doubt, as we all realize, that a new era of oral hypoglycemic therapy is here. There is every indication that, if these drugs are carefully used, a great deal of comfort and help can be given to many properly selected patients. It is fortunate that a large number of experienced people have contributed to this monograph in appraising the value of chlorpropamide.

There are a few papers that relate to insulin alone; this raises the total to fifty thousand plus a few since 1921. Robert Williams* has given us a comprehensive discussion of the destruction of insulin by homogenates and fractions of many tissues. The activity in tissues varies from the maximum in liver to the minimum in blood plasma. Williams makes the distinction between the reductive and proteolytic mechanisms of degradation (glucagon is destroyed only proteolytically). Little change in the insulin-destroying activity was noted after infective and traumatic insults. Traumatization of rat muscle *in vivo* did increase the insulin-destroying effect. Williams paid eloquent tribute to Mirsky's* original contributions in this field. In discussion Mirsky emphasized that, in referring to insulinase, he envisaged a labile saturated proteolytic system. Both Williams and his colleagues and Mirsky have now prepared a highly purified insulinase, and it is obvious that studies of the physiological role of this enzyme will be greatly facilitated.

Prout and Evans* have measured the *in vivo* disappearance of I^{131} insulin and feel that the fixation of the hormone by tissues is not a significant factor in this removal. The intact insulin is distributed in a volume similar to that of extracellular water, that is, it is distributed throughout the extracellular space.

There are many problems on which we all seek further information. What is the mechanism of action of any of the oral hypoglycemic agents? What evidence is there that an increased secretion or liberation of insulin is produced? Loubatières provided our first evidence in a classically physiological way, that is, by removing the pancreas and noting the disappearance of the effect. Houssay* and many other authors have confirmed this. Most of

* Elsewhere in this monograph.

the recent data have been obtained in studies on tolbutamide and the situation is still unsettled. I shall not attempt to review here all the direct and indirect evidence that has been obtained in a variety of ways. There have been studies on the morphological changes of the beta cells[10-17] and on the amounts of insulin extractable from the pancreas.[17, 18] The direct attack—that is, estimation of the insulin content of plasma—has many supporters.[19-25] Indirect approaches have utilized cross circulation tests (Loubatières,[26] Foà,[27] Colwell[28-29]), perfusion experiments, eviscerated preparations,[30-32] and a great variety of metabolic studies—glucose tolerance tests[33-37] and estimations of such factors as serum inorganic phosphate,[34-36, 39, 40] potassium,[37, 40] nitrogen balance,[37, 41] blood pyruvate,[36, 37, 43, 44] and lactate,[36, 47, 44] plasma UFA,[27, 45] ketones,[2, 42] and the RQ.[23, 41, 46] The effect on the decrease in sugar output by the liver[34, 47-49] sometimes has been used, in effect, as a test for insulin, and changes in enzyme activity in the liver obviously might be similarly interpreted[50] in certain cases.

The importance of knowing the extent and duration of insulin liberation produced by the various oral hypoglycemic agents cannot be overemphasized. How can we know to what extent insulin itself changes enzyme activity in the liver and how much direct action a drug is exerting without this information? The valuable studies of Sass and Handelsman* on transaminases in liver homogenates of animals that have received chlorpropamide illustrate this point. Indirect approaches can help, of course. The situation is similar when we consider sugar production by the liver, as I shall point out later.

I shall not attempt to give an appraisal of all the indirect evidence that is very strongly in favor of some insulin liberation after administration of the oral agents. Positive direct evidence has been reported by von Holt,[19] Goetz,[2?] and Pfeiffer *et al.*[25] The methods used are not above criticism by the *investigators themselves as well as by others*. *Recently*, Renold (*personal* communication) and his group were unable, even with their very sensitive and reliable method, to detect in man any increase in plasma insulinlike substances after administration of the oral agents. They do find an increase in the *amount of these materials* in pancreatic vein blood of the dog. A survey of this field leads to the conclusion that one aspect of the action of the hypoglycemic oral agents is almost certainly the liberation of a certain amount of extra insulin, but the possibility remains that the mere presence of a little insulin furnishes the essential link.

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have sup-

ported this conclusion In human subjects he found that a slow infusion of insulin produced a decreased hepatic output of glucose, but no indication of increased peripheral use Larger doses of insulin gave both effects

Madison, Unger, and Kaplan* found in human subjects that slow infusions of glucagon-free insulin, which produced a good fall in arterial blood sugar, caused no change in arteriovenous difference Tolbutamide produced the same effect They reported that in dogs with portacaval fistulas the administration of insulin consistently produced a decreased hepatic output of sugar These workers emphasized the various differences between endogenous and exogenous insulin The only established difference between natural and exogenous insulin relates to the route of entry of the hormone We all know that depancreatized dogs and some severe diabetic patients have lived a life time (that is, six years and thirty seven years, respectively) on exogenous insulin

Schambye and Tarding* found that tolbutamide produced an inhibition of hepatic glucose output as judged by plateauing of the curve of specific activity of C^{14} glucose in normal and depancreatized dogs and in diabetic patients The inhibition in the normal organism lasted no longer than 40 min but in the diabetic it lasted at least 2 hours Although insulin given intravenously and intraportally in normal dogs could not be found to decrease hepatic output of sugar, tolbutamide did produce this effect These differences in the observed effects of insulin emphasized again the necessity of attempting to standardize our tools—the type and amount of insulin, for example and many other variables

I propose now to emphasize a point or two on the effect of insulin on the liver Perfusion of the isolated liver (Miller, Hamff*) has demonstrated a rather prompt effect of insulin on fat formation, and I shall not be surprised to see revealed a prompt, direct action on sugar and amino acids in this tissue The fundamental interest and practical importance of this subject must be obvious We cannot readily investigate the *direct* action of the oral agents on liver if these agents liberate insulin which, in turn, affects the liver There is, of course, no doubt about many very important delayed and perhaps indirect actions of insulin on the liver De Duve[31] and his colleagues recently discussed this matter at the International Diabetes Federation in Dusseldorf Germany While I feel that small amounts of insulin are probably liberated or, at least, are necessary for the action of the oral agents, I must stress the fact that the exploration of the direct effect of these agents on the liver has hardly been started Downie* has aroused our interest by reference to Bornstein's[32] work on the effect of chlorpropamide on the oxidation of C^{14}-labeled alanine to CO_2 in liver slices Mirsky* has stimulated us again with his finding that tolbutamide prevents the diabetogenic action of somatotropin in dogs Zarowitz* compared the glucose and tolbutamide *tolerance tests in nondiabetic and diabetic patients He feels that the tol*butamide test detects some mild diabetics that the older test would not reveal Tolstoi* did not see the advantage of and justification for the tolbutamide test

However, we are concerned particularly in this monograph with chlor-

* Elsewhere in this monograph

propamide It is obvious that this compound is an effective agent and that, under certain circumstances, it can be given safely for prolonged periods to humans

McLamore's work* gives us a great deal of information on the relation of chemical structure to hypoglycemic action Schneider† and Mary Root,‡ agreed on the greater activity of chlorpropamide as compared to tolbutamide in mice, rats, and dogs The former was slightly more toxic Chlorpropamide was active in monkeys

In rats there was a degranulation of beta cells, no effect in alloxan diabetes, a *positive effect in hepatectomized preparations* and an *enhanced effect in* adrenalectomized animals Schneider reported certain toxic effects (predominately on the CNS) with large doses of chlorpropamide in dogs, but he stated that 120 mg kg was well tolerated for 20 months There was no change in various liver function tests, this is in contrast to the findings of A Sirek and O Sirek[53] in our laboratory with tolbutamide which, at a level of 30 mg/kg, does produce hepatic injury in normal and diabetic dogs We should test chlorpropamide under the same conditions, using dosages that produce the same hypoglycemic effects The liver lesions seen in a few patients receiving chlorpropamide resemble strikingly those seen in dogs given tolbutamide, but the finding in dogs must not be ' transferred" to human subjects, the striking absence of liver lesions in many, many thousands of patients receiving tolbutamide must be emphasized again

Lundbaeck* and Clarke* differ in their finding of glucose uptake by diaphragm in *in vitro* studies Lundbaeck has reported an increased carbohydrate tolerance in patients given chlorpropamide or tolbutamide *Danowski** has studied chlorpropamide in carefully standardized normal and diabetic subjects He feels that the first effect may be to decrease gluconeogenesis, the second, perhaps, to increase insulin liberation He studied particularly blood glucose and phosphate detecting no change in serum inorganic phosphate in diabetics given chlorpropamide His report on the increased immediate utilization of sugar and the fall in serum phosphate after cortisone is intensely stimulating

On the clinical use of chlorpropamide, all agree, as Tolstoi summarized, that chlorpropamide is an effective agent, and one more potent than tolbutamide As Beaser emphasized, the drug should be criticized only for its [illegible] the m[illegible]mal effective dose [illegible] blood level and [illegible]amide the same effects were produced Craig* agreed but others have been less certain of the significance of this relationship *Kelly West* feels that chlorpropamide* is more potent intrinsically, but he has not as yet measured blood levels Beaser, using different doses of chlorpropamide, found a direct relationship between dose and blood level, but not necessarily between dose and blood sugar values Hamff* reported three cases of jaundice with the use of large

* Elsewhere in this monograph
† Of Chas Pfizer & Co Inc Brooklyn N Y
‡ Of Eli Lilly and Company Indianapolis, Ind

doses of chlorpropamide Two of these patients later did well on smaller doses and the third was transferred to tolbutamide

There have been primary and secondary failures with both drugs and, in certain cases, the other drug was effective Nabarro's* statement on the incidence of acquired resistance to tolbutamide was noteworthy, as many as 14 per cent of his cases showing resistance in 18 months Insulin consistently has been found to be effective, and its dose can safely be increased when necessary

The selection of cases for oral therapy is a vitally important matter Duncan,* Hamwi,* and many others have discussed this problem Ketosis on withdrawal of insulin is considered to contraindicate the use of the oral agents In addition to careful selection of patients, Duncan emphasized that these drugs should control diabetes at least as well as insulin in mild cases and, if used with insulin in the more severe ones dramatic evidence of their effectiveness should be available

I was very interested in Stowers * paper on competition for tubular excretion between chlorpropamide and salicylates Salicylates raised the level of chlorpropamide in plasma, this finding may have a bearing on Segarra s* work in tuberculous patients who were receiving both drugs

Henry Ricketts* stressed the necessity for a uniform method of clinical investigation when drugs are to be compared All the established principles of good treatment of diabetes must be followed before a new variable is introduced

The precise incidence of the complications produced by chlorpropamide in therapeutic doses is not known It must be emphasized that each complication should be most thoroughly investigated The evaluation of the significance of complications in the older age group is, of course particularly difficult

Concluding Remarks

I have approached this subject as a physiologist and not as a clinician We have in chlorpropamide an effective agent that appears to be more potent than the two drugs previously used Adequate dose response curves are not yet available Chlorpropamide is longer acting than tolbutamide and can be given in smaller doses, which should always be small and should never exceed 750 mg per day It is probable that 500 mg or less would be adequate in most patients, and this amount would be safer The safe therapeutic range of chlorpropam[illegible]

to this point

be determined

high levels and small increases in dose may have a surprisingly great effect on blood levels Chlorpropamide tends to accumulate, and hypoglycemia may be disturbing, particularly during the early period of adjustment Patients should be watched very carefully, since delayed responses inde-

* Elsewhere in this monograph

and is illustrated again by a case presented by Tolstoi Wherever possible patients should be standardized on diet alone If more specific therapy is needed the insulin requirement of all patients should be determined If an oral agent is then used its insulin equivalent is thus made available These are valuable data and they should be obtained whenever possible

Someone has observed that the patients who respond best to the oral agents are those who do not need them There is, of course, a real need for these agents in the right cases but it is becoming increasingly obvious that many patients are receiving these drugs who could and should be controlled by diet alone As Dobson said the oral agents are frequently a convenience and not a necessity In ten years we shall know if this convenience will have been perfectly safe

I have attempted to quote a number of the distinguished clinicians and I conclude by referring to the finding made by Alexander Marble* that chlorpropamide is an effective agent Marble encountered a disturbing side effect in one patient of seventy two even with a relatively small dose of the drug He believes as I do that there are many advantages in having a *number of safe active oral agents available and he stressed the fact that* more exact clinical work on chlorpropamide is needed to determine its proper sphere of usefulness

It is important that the national diabetes associations intensify their efforts at the present time to help physicians in the selection of cases and in development of an appreciation of the risks involved and the benefits to be obtained from the oral hypoglycemic agents

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